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Loews Annapolis, January 7-10 2014



"...and uphill both ways"



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On the cover

Vincent D'Amico for providing the cover artwork, "...and uphill both ways"



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25TH USDA INTERAGENCY RESEARCH FORUM ON INVASIVE SPECIES

**January 7-10, 2014
Loews Annapolis, Maryland**

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FOREWARD

This meeting was the 25th in a series of annual USDA Interagency Research Forums that are sponsored by the Forest Service, Animal and Plant Health Inspection Service, National Institute of Food and Agriculture, and Agriculture Research Service. The Group's original goal of fostering communication and providing a forum for the overview of ongoing research among the Agencies and their cooperators is being realized and facilitated through this meeting. This proceeding documents the efforts of many individuals: those who organized and sponsored the meeting, those who provided oral and poster presentations, and those who compiled and edited the contributions. The proceedings illustrates the depth and breadth of studies being supported by the agencies and their many cooperators and demonstrates the benefits and accomplishments that can result through the spirit of collaboration.

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CONTENTS

PRESENTATIONS.....	1
KEYNOTE ADDRESS: INVASION BIOLOGY - WHERE DID IT COME FROM, WHERE IS IT GOING, AND WHY ARE SOME PEOPLE TRYING TO KILL IT?	
by David Simberloff	2
RESEARCH REPORTS	4
INVASION DYNAMICS OF THE MOUNTAIN PINE BEETLE IN JACK PINE FORESTS: ROLES OF HOST SECONDARY COMPOUNDS by Nadir Erbilgin.....	5
RECENT SHIFTS IN THE POPULATION DYNAMICS OF TWO SAWFLY SPECIES: THE IMPACT OF CLIMATE CHANGE? by Christa Schafellner, Martina Marschnig, Martin Schebeck, and Axel Schopf	6
DETECTION OF ASIAN WOOD BORING BEETLES IN HIGH-RISK SITES: TWO CASE STUDIES FROM ITALY by Davide Rassati, Lorenzo Marini, Massimo Faccoli, Edoardo Petrucco Toffolo and Andrea Battisti	9
NANOFABRICATED DECOYS USED IN AN ELECTROCUTION TRAP FOR EMERALD ASH BORER AND OTHER <i>AGRILUS</i> SPP. by Michael J. Domingue, Drew P. Pulsifer, Akhlesh Lakhtakia, Victor C. Mastro, and Thomas C. Baker	11
OUT OF THE CITIES AND INTO THE FOREST: RECENT DETECTIONS IN URBAN FOREST OF BRITISH COLUMBIA by Leland M. Humble.....	13
CLASSICAL BIOCONTROL OF EMERALD ASH BORER IN NORTH AMERICA: MONITORING DEMONSTRATES SUCCESSFUL ESTABLISHMENT by Leah S. Bauer, Jian J. Duan, Juli R. Gould, Kristopher J. Abell, Jonathan P. Lelito, and Roy Van Driesche	15
UNEXPECTED RAPID APPEARANCE AND SPREAD OF <i>ENTOMOPHAGA MAIMAIGA</i> IN CENTRAL-EASTERN EUROPEAN GYPSY MOTH POPULATIONS by Gy. Csóka, M. Zubrik, B. Hrasovec, M. Tabakovic-Tosic, A. Kunca, A. Hirka, M. Pernek, M. Glavendekic, M. Dautabasic, G. Georgieva, A. Hajek and D. Pilarska	18
SYNTHESIS OF RESEARCH ON <i>SIREX NOCTILIO</i> by Ann E. Hajek, E. Erin Morris, Louela Castrillo, Stefanie A. Kroll, Mark A. Sarvary, Miriam F. Cooperband, David Williams	20
INVASION BIOLOGY OF <i>PHYTOPHTHORA CINNAMOMI</i> IN EASTERN US OAK FORESTS: ITS CURRENT AND POSSIBLE IMPACT UNDER PROJECTED CLIMATE CHANGE by Yilmaz Balci.....	23
CLASSICAL BIOCONTROL OF EMERALD ASH BORER: A LIFE TABLE ANALYSIS OF POPULATION IMPACTS OF INTRODUCED NATURAL ENEMIES by Jian J. Duan, Kristopher J. Abell, Leah S. Bauer, Juli R. Gould and Roy Van Driesche ...	24
MITIGATING PATHWAYS FOR FOREST PESTS	26
EVALUATION OF PEST REDUCTION AFTER IMPLEMENTATION OF ISPM 15 by Robert Haack	27

URBAN FORESTS: THE FRONT LINE FOR INVASIVE SPECIES	30
BELOWGROUND CAUSES AND CONSEQUENCE OF EUROPEAN BUCKTHORN INVASIONS INTO REMNANT URBAN WOODLANDS by Basil V. Iannone III, David H. Wise, Liam Heneghan, Christopher Oswalt, Sonja Oswalt, Nathan Clause, Bryan C. Pijanowski, and Songlin Fei	31
DETECTING THE NEXT WORM IN THE “BIG APPLE” by Richard Hallett and Nancy Falxa-Raymond	33
NORTHEAST BIOCONTROL REGIONAL PROJECT (NE-1332): UPDATES ON REGIONAL BIOCONTROL PROJECTS.....	37
INTERACTIONS BETWEEN NATIVE PREDATORS AND <i>CYZENIS ALBICANS</i> IN THE CONTROL OF WINTER MOTH (<i>OPEROPHTERA BRUMATA</i>) IN THE NORTHEASTERN US by Hannah J. Broadley and Joseph S. Elkinton	38
RECENT ADVANCES IN QUARANTINE MONITORING AND MANAGEMENT OF EXOTIC WOOD BORING INSECTS	39
FIELD TESTING OF GENERIC PHEROMONE LURES FOR CERAMBYCIDS IN CHINA by Jacob D. Wickham, Rhett D. Harrison, Wen Lu, Yi Chen, Jocelyn G. Millar and Lawrence M. Hanks	40
EARLY DETECTION OF CERAMBYCIDAE AND CURCULIONIDAE SPECIES IN SURVEY TRAPS: WHAT ARE WE MISSING? by Jon Sweeney, Peter J. Silk, Reggie Webster, Rob Johns, Peter Mayo, Troy Kimoto and Vic Mastro	42
ENHANCING IDENTIFICATION OF CERAMBYCIDS AND BUPRESTIDS FROM PORT INTERCEPTIONS IN SOLID WOOD PACKING MATERIAL by Peter Reagel, Ann Ray, Scott Myers and Hannah Nadel	43
CERAMBYCID INTERCEPTIONS AND THEIR IMPACT IN THE NETHERLANDS? by Brigitta Wessels-Berk	44
CERAMBYCIDS OF QUARANTINE CONCERN FOR EPPO REGION by Andrei D. Orlinski	45
BARCODING THE CERAMBYCIDAE: PROGRESS IN THE MOLECULAR IDENTIFICATION OF LONG-HORNED WOODBORERS by Leland M. Humble	47
POSTERS.....	48
DETECTION OF ALIEN WOOD BORING BEETLES IN HIGH-RISK SITES: TWO CASE STUDIES FROM ITALY by Davide Rassati, Lorenzo Marini, Massimo Faccoli, Edoardo Petrucco Toffolo and Andrea Battisti.....	49
OPTIMIZING RESOURCE ALLOCATION BY INTEGRATING PATHWAY ANALYSES AND ECONOMIC IMPACTS by Kevin M. Biggsby, D.A. Burnie and D. Christie.....	51
DROUGHT AS A PREDISPOSING FACTOR FOR BARK BEETLE ATTACK by S. Netherer, E. Blackwell, P. Henschke, B. Matthews, P. Hietz, K. Katzensteiner, S. Kikuta, J. Pennerstorfer, S. Rosner, H. Schume and A. Schopf.	52
<i>SIREX NOCTILIO</i> (HYMENOPTERA: SIRICIDAE) MATING BEHAVIOR by Isis A. L. Caetano and Ann E. Hajek	54
CONTAINING DOMESTIC SPREAD OF PESTS: HOW TO OVERCOME THE CHALLENGES? by Faith T. Campbell	55
THE BLACK OAK GALL WASP ON CAPE COD AND MARTHA’S VINEYARD by Monica Davis and Joe Elkinton.....	56
BUYING TIME: POTENTIAL ROLE OF BIOCONTROL IN THE RECOVERY OF NATIVE VEGETATION AFTER A PEST INVASION by Elan Margulies, Inés Ibáñez, Leah Bauer, Jian Duan and Roy Van Driesche	57
INVESTIGATING THE EFFECTS OF MATERNAL IMMUNE PRIMING ON THE EFFICACY OF A FUNGAL BIOLOGICAL CONTROL AGENT by Joanna J. Fisher and Ann E. Hajek	58

CASCADE EFFECT OF LAUREL WILT DISEASE ON THE PALAMEDES SWALLOWTAIL BUTTERFLY IN THE SOUTHEASTERN UNITED STATES by John Formby, Kelly L. Oten and John J. Riggins.....	59
CANARIES IN THE COALMINE: URBAN WARMING PREDICTS CLIMATE CHANGE EFFECTS ON FOREST PESTS by Steven D. Frank, Elsa K. Youngstead, Adam G. Dale, Emily K. Meineke and Robert R. Dunn	60
EFFECTS OF URBAN FOREST FRAGMENTATION ON ABUNDANCE, DIVERSITY AND HEALTH OF NATIVE BEES by David Gardner and Deborah Delaney	62
MICROSCLEROTIA APPLIED IN HYDROMULCH TO CONTROL ASIAN LONGHORNED BEETLES by Tarryn A. Goble, Ann E. Hajek, Mark Jackson and Sana Gardescu	63
ALLIANCE FOR SAVING THREATENED FORESTS: BREEDING FOR RESISTANCE TO HEMLOCK WOOLLY ADELGIDS by Ben Smith, Fred Hain and John Frampton	64
LOCAL CONDITIONS INFLUENCE GYPSY MOTH OUTBREAK REGULATION BY <i>ENTOMOPHAGA MAIMAIGA</i> by Ann E. Hajek, Patrick C. Tobin and Kyle Haynes	65
NATIVE AND NON-NATIVE CERAMBYCID BEETLES IN URBAN FOREST FRAGMENTS by Kaitlin Handley, Judith Hough-Goldstein, Meg Ballard, Larry Hanks, Jocelyn Millar and Vincent D'Amico III	66
CREATION AND VALIDATION OF A DEGREE-DAY MODEL FOR <i>SIREX NIGRICORNIS</i> F. (HYMENOPTERA: SIRICIDAE) IN ARKANSAS by J.A. Hartshorn, L.D. Galligan, D.J. Dalzotto and F.M. Stephen.....	67
SEX-SPECIFIC TRAIL PHEROMONE OF THE ASIAN LONGHORNED BEETLE, <i>ANOPLOPHORA GLABRIPENNIS</i> by Kelli Hoover, Melody Keena, Maya Nehme, Peter Meng, and Aijun Zhang.....	68
BIOLOGICAL CONTROL OF EMERALD ASH BORER IN MARYLAND: AN UPDATE ON PARASITOID RECOVERY AND ESTABLISHMENT by David E. Jennings, Jian J. Duan, Dick A. Bean, Philip B. Taylor, Kimberley A. Rice, Charles W. Pickett, Steven K. Bell and Paula M. Shrewsbury	69
EFFECTS OF TEMPERATURE ON NEONATE <i>LYMANTRIA</i> SURVIVAL WITH AND WITHOUT FOOD by Melody A. Keena and Juan Shi	70
FIELD OBSERVATIONS OF <i>SIREX NOCTILIO</i> MATING AND MALE AGGREGATION BEHAVIOR by Flora Krivak-Tetley, Katalin Böröczky, Marc Bouwer, Jeff Garnas, Andrew Liehold and Bernard Slippers	71
<i>SCYMNUS CAMPTODROMUS</i> DEVELOPMENT AND PREDATION OF HEMLOCK WOOLLY ADELGID by Samita Limbu, Melody Keena, David Long and Kelli Hoover	72
ATTRACTION OF NON-CERAMBYCIDS TO CERAMBYCID PHEROMONES IN SOUTHEASTERN USA by D.R. Miller, C.M. Crow, J.D. Sweeney, P. Mayo and P.J. Silk	73
VARIATION IN EFFECTS OF CONOPHTHORIN ON ATTRACTION OF AMBOSIA BEETLES TO ETHANOL-BAITED TRAPS by D.R. Miller, C.M. Crow, K.J. Dodds, E.R. Hoebeke, T.M. Poland, R.J. Rabaglia and E.A. Willhite	74
DROUGHT AS A PREDISPOSING FACTOR FOR BARK BEETLE ATTACK by S. Netherer, E. Blackwell, P. Henschke, B. Matthews, P. Hietz, K. Katzensteiner, S. Kikuta, J. Pennerstorfer, S. Rosner, H. Schume and A. Schopf.....	75
CONSERVATION OF ASH DIVERSITY: EFFECTS OF WIDE-SCALE INSECTICIDE APPLICATION ON GENETIC STRUCTURE, RECRUITMENT, AND REGENERATION OF ASH IN FORESTS INVADDED BY EMERALD ASH BORER by Erin M. O'Brien and Daniel A. Herms	77
ASH MORTALITY CAUSED BY EMERALD ASH BORER IMPACTS FOREST FLOOR INVERTEBRATE COMMUNITIES by Kayla I. Perry, Rosemary K. Walker and Daniel A. Herms	78
THE US ASIAN GYPSY MOTH PEST EXCLUSION PROGRAM: MONITORING AND SHIP INSPECTION PROGRAMS IN JAPAN, KOREA, CHINA AND THE RUSSIAN FAR EAST by Baode Wang, Victor C. Mastro, Michael Simon, D.A. Steve Munson and Dean Duval.....	79

RECENT OUTBREAKS OF THE APPLE JEWEL BEETLE (COLEOPTERA: BUPRESTIDAE) IN NORTHWEST CHINA: A POTENTIAL RISK TO APPLE PRODUCTION by Xiao-Yi Wang, Zhong-Qi Yang, Zhi-Yong Wang, Liang-Ming Cao and Jian J. Duan	81
EMERALD ASH BORER RISK ASSESSMENT AND SAMPLING DESIGN by John Withrow, Ian Leinwand, Marla Downing, Frank Sapio, Paul Chaloux and Joe Beckwith	83
ATTENDEES.....	84

PRESENTATIONS

Keynote Address

Invasion biology – Where did it come from, where is it going, and why are some people trying to kill it?

David Simberloff

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ABSTRACT

Invasion biology is a new science; people recorded introduced species beginning in the 18th century but until recently focused almost wholly on biogeography – which species are where – rather than impacts. Contrary to popular belief, Elton's 1958 monograph did not originate modern invasion biology. Elton focused largely on impacts and adumbrated many of the concerns of modern invasion biology, but his book had little influence following its publication. Rather, the founding event was the Scientific Committee on Problems of the Environment (SCOPE) project of the 1980s, spearheaded by Harold Mooney, which produced several widely read books. Invasion biology is so young that the SCOPE questions are still relevant: (1) Why are some species invasive? (2) Why are some habitats invulnerable? (3) How can knowledge about (1) and (2) aid management of troublesome invaders? The questions have evolved, and we now realize that questions (1) and (2) are inextricably linked, but the kinds of detailed, case-specific research that have led to this understanding remain important.

One of the newer growing points of invasion biology is the study of invasions that affect entire ecosystems. Although Peter Vitousek called for research on such invasions during the SCOPE projects, such studies remained a minor component of modern invasion biology until recently. They have now become one of its leading edges, particularly as ecological research on aboveground-belowground interactions has proliferated. This research has revealed how invaders that modify nutrient cycles, hydrological or fire regimes, and physical structures can impact entire ecosystems. Although several evolutionists participated in the SCOPE project, evolutionary research was not part of the SCOPE agenda and did not become a major part of modern invasion biology until the last decade. However, spurred by the explosive development of accessible and inexpensive tools of molecular genetics, evolutionary research now plays an increasingly prominent role in invasion biology. In addition to increasing evidence of rapid evolution of both invaders in their new ranges and native species in response to invaders, the burst of evolutionary research has shown that many invasions previously thought to have arisen from single introductions instead arose from several introduction events, bringing increased genetic diversity. This finding helps to resolve the “genetic paradox of invasion biology,” the apparent ability of populations initiated from small (and presumably genetically depauperate) founder populations nevertheless to grow to robust and ecologically dominant components of the biotic community. Multiple introductions sometimes boost the amount of genetic variation.

In the midst of this burst of research on invasions and increasing understanding of their impacts, critics have lambasted invasion biology and management on several grounds, claiming that the publicized harmful impacts are overblown, that invasions often increase biodiversity, that attempts to control invasions are futile, and that the entire enterprise is tainted with xenophobia.

The charge that invasion impacts are exaggerated is based on the widely cited belief that the great majority of introduced species have no substantial impact – Mark Williamson's “tens rule” is an early avatar of this view. However, the great majority of introduced species have not been studied in their new ranges, so we cannot conclude that their impacts are negligible. As invasion biology has grown since its inception, we have learned that many invaders assumed to be innocuous instead substantially affect native species, communi-

ties, and ecosystems. It has also become clear that some sorts of invasion impacts, though subtle and not obvious without detailed research, are nonetheless consequential – changes in the nitrogen cycle effected by invasive nitrogen fixers is an example. It is also apparent that some introduced species that are initially restricted to small, insignificant populations near their site of arrival – sometimes for decades – relatively suddenly explode across the landscape and become damaging invaders. Sometimes the cause of the lag and its sudden termination can be easily deduced – for instance, the arrival of an important mutualist, like a pollinator or a mycorrhizal fungus for a plant. Other times the lag and its ending are mysterious. However, the phenomenon cannot be doubted, and this means that we cannot assume an invasion will remain harmless even if it seems so today. The charge that the threat of non-native species is overblown is often accompanied by one or both of two observations: that some native species become “invasive,” and that some non-native species benefit some native species and communities. Both of these observations are true, but they should be taken with a grain of salt. Recent research shows that invasive behavior by native species is far less common than that by non-native species, and when it occurs, it is almost always in the wake of some other anthropogenic change, such as modified fire cycle or overgrazing. Many of the supposedly beneficial non-native invaders also have harmful detrimental impacts that would seem to far outweigh any conservation benefit they might confer.

Invasions do sometimes increase species richness locally, but globally and regionally they have decreased species richness. Whether one is gratified more by the local increase than by the global decrease is a matter of taste, or perhaps religion. An example of the issue is the loss of 55 endemic land and fresh-water birds from the Hawaiian Archipelago in the wake of human colonization. This loss is approximately countered by the establishment of 53 introduced bird species. However, the endemic species are extinguished globally, whereas all but one of the 53 introduced species is common in its native range, and many have been widely introduced elsewhere. Further, by vectoring avian pox and malaria, they contribute to the endangerment of the remaining native Hawaiian birds.

The charge of xenophobia has been raised primarily by persons outside the life sciences – philosophers, historians, anthropologists, sociologists. In a classic social construction of a science, they largely ignore evidence on impact of targeted invasive species and present no evidence that it is xenophobia, rather than impacts, that motivates research and management efforts.

The argument that efforts to stem the flow of introduced species and eradicate or manage those that establish are futile ignores a number of major success stories in both eradication and maintenance managements. Major advances abound in technologies of eradication, chemical control, biological control, and physical or mechanical control, as well as more specialized approaches such as sterile male technique and pheromonal mating disruption. Radically new approaches appear occasionally. For instance, the resurgence of interest in genetically based autocidal techniques in the era of transgenes has already produced successful field tests for fishes and dipterans.

The above arguments do not appear to have affected policy on and management of introduced species. Indeed management efforts have increased, surely in response to increased recognition of the cost of invasions. No doubt continued education of the public and its policymakers about impacts and management of introduced species will vitiate the force of these arguments. However, maintenance management and eradication projects targeting invasive mammals and birds frequently confront objections from advocates of animal rights. These objections will not easily be countered, as they are based on deep-seated philosophical or religious views on the relative weight of the rights of individual sentient animals and the rights of species.

PRESENTATIONS
Research Reports

Invasion Dynamics of the Mountain Pine Beetle in Jack Pine Forests: Roles of Host Secondary Compounds

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ABSTRACT

Host plant-herbivore interaction is one of the most challenging issues in invasion biology and has cascading impacts on establishment success of invasive herbivore populations, their population dynamics and potential for invasiveness within their novel range. My group investigated whether a dominant host tree species is suitable for the invasion success of the mountain pine beetle, (*Dendroctonus ponderosae*), in the evolutionarily naïve jack pine forest ecosystem (in terms of encounters with mountain pine beetle) in western North America. Since pheromone communication regulates successful host plant colonization by bark beetles and plant secondary chemicals can influence the production and release of bark beetle pheromones we examined the role of secondary chemistry of historical (lodgepole pine) and novel (jack pine) host trees on pheromone production and host colonization by mountain pine beetle. We evaluated the pheromones emitted by beetles feeding on both host species and found that beetles emit pheromone compounds from the novel host similar to those emitted from its ancestral host, but in larger quantity. The amount of *trans*-verbenol, female aggregation pheromone, released was more in the novel host. Likewise the release of one host monoterpene, 3-carene, was higher in jack pine. We developed release devices to reflect the differences in pheromone and 3-carene emission between host species and tested them in a field experiment. While all baits attracted more beetles than the blank control, there was no difference in beetle capture in traps baited with the chemicals mimicking the two host tree species. Addition of 3-carene to the pheromone mix improved the beetle attraction. Furthermore, broods that emerged from the novel host were superior in terms of fat content and capacity to fly compared to broods that emerged from the ancestral host. These results support previous predictions that there seem to be no biological impediments for the establishment and spread of mountain pine beetle in jack pine boreal forest.

Recent Shifts in the Population Dynamics of Two Sawfly Species: the Impact of Climate Change?

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ABSTRACT

Secondary spruce forests in the Alpine foothills of Austria and Bavaria (Germany) have a long history of sawfly infestations (Sedlacek, 1904; Schimitschek, 1935; Führer and Nopp, 2001). Severe outbreaks of the little spruce sawfly, *Pristiphora abietina* (Christ.) (Hymenoptera: Tenthredinidae) mainly affected lowland Norway spruce (*Picea abies* [L.]) stands, while high population densities of the mountain spruce sawfly, *Pachynematus montanus* (Zaddach) (Hymenoptera: Tenthredinidae), were restricted to pure spruce stands at altitudes above 800 m (Wiener, 1990).

The two sawfly species have similar life histories; adults eclose soon after spruce bud burst and oviposit eggs singly in slits cut in the needles of new flushing shoots. Young *P. abietina* larvae feed exclusively on current-year foliage through four (male) or five (female) instars; larvae of *P. montanus* feed first on new needles, from instar three larvae consume older age classes of needles. Finally, mature larvae drop to the ground and spin a cocoon in the upper duff layer, where they overwinter as prepupae (eonymph or pronymph). A number of cocooned larvae, sometimes higher than 50 % within local populations, may remain in prolonged diapause for two or more winters (Schedl, 1991).

Repeated infestations by *P. abietina* cause dieback of the leaders and a bushy deformation of the crown; losses of height and increment growth can be considerable, but the trees do not die. Despite significant year-to-year fluctuations, the long-range oscillations in population dynamics are concurrent among all populations of the affected areas. A number of abiotic and biotic factors determines the oscillatory pattern of population fluctuations (e.g., coincidence of budburst and sawfly emergence in spring, weather conditions during larval feeding, food shortage, parasitism, predation, pathogen infection) (Ohnesorge, 1957; Ohnesorge and Thalenhorst, 1966).

In 2010/11 significant defoliation was reported from lowland spruce plantations in the Northern Flachgau (Salzburg, Austria) and in parts of the pre-Alpine area of Upper Austria, regions with regular outbreaks of *P. abietina* during the past four decades (Kurir, 1982). Close inspections of the affected sites in Austria and adjacent areas in Bavaria, however, revealed that the recent damage was not caused by *P. abietina*, but instead by *P. montanus* which has never reached outbreaking levels at these locations before (Egginger, 2013). Subsequently, heavily infested spruce trees were attacked by the European spruce bark beetle, *Ips typographus* (L.) (Coleoptera, Curculionidae) which then killed the trees.

In 2012 we started the project 'Forest pests and climate change', funded by the European Union (Interreg J00337), in order to investigate this unprecedented outbreak of the mountain spruce sawfly in lowlands of the foothills of the European Alps. One major question is if and how climate change influences the coincidence of sawfly swarming and budbreak of spruce at different elevations. A shift in the phenolo-

gy of one or the other sawfly species relative to their host plant during the last years had eventually allowed *P. montanus* to out-compete *P. abietina* at low elevations.

This study summarizes results from several low elevation (<450 m a.s.l.) monitoring sites in Austria and Bavaria on sawfly population densities, phenology, and cocoon mortality and records feeding damage and tree defenses of infested trees analyzed in 2013.

Cocoons were not evenly distributed among the host trees, but cocoon densities in the duff layer were extraordinarily high, ranging from 10.000 to 30.000 per square meter. About 60 to 80 % of these were *P. abietina* cocoons and 16 to 33 % were cocoons of *P. montanus*. The high numbers reflect an accumulating effect over years resulting from the excellent mechanical and chemical stability of the cocoon silk proteins under environmental conditions. Based on characteristic appearance and holes in cocoons (described by Nägeli, 1936), cocoons were scored for successful adult sawfly emergence or the likely source of mortality (predation, parasitism, desiccation, fungal infection). An estimated 30 % of the sawflies were able to emerge, while mortality attributed to natural enemies was extremely high. Parasitoids killed approximately 50 % of the cocoons and predators killed up to 10 %. Desiccated larvae were observed in 10 % of all cocoons, while disease was evident less frequently.

Intact cocoons were separated from those with holes and dissected. *P. abietina* cocoons did not contain living unemerged sawflies, but instead were empty or contained dead and dried nymphs (due to fungi and other unidentified causes). Approximately 10 % of intact *P. montanus* cocoons contained living eo- or pronymphs, suggesting that they were 1-2 years old.

Adult sawfly emergence in spring 2013 was recorded with photo-electors. Almost 95 % of the captured sawflies were *P. montanus*, while the rest belonged to unidentified *Pachynematus* species and *Cephalcia* sp. No *P. abietina* wasps were trapped. The swarming period of *P. montanus* started in late April, most adults emerged in the first and second week of May. The sex ratio was 1:1, indicating the retrogradation phase of the recent sawfly outbreak.

When the mature larvae dropped to the ground in late June, they were captured in funnel traps connected to boxes filled with needle litter and duff. We collected on average 188 cocoons per square meter crown cover, 185 were *P. montanus* (i.e. 98 %), two *P. abietina* cocoons and one cocoon of an unidentified *Pachynematus* species. In October, half of the *P. montanus* cocoons contained dead sawflies and half contained living pronymphs (65 %) or eonymphs (35 %). Parasitoids and fungi killed each 10 % of the sawflies, while desiccated larvae were observed in the vast majority of the cocoons with dead sawflies.

Eighteen *P. abies* trees (80 to 100 years old) were selected from three study plots. From each two trees with high, medium and no visible feeding injuries, two branches from the 7th and 14th whorl were randomly sampled in summer 2013 and the mean annual growth increments and needle losses due to sawfly feeding were recorded. Criteria for categorizing damage classes (I to IV) were based on the percentage of needle loss relative to control branches with no feeding damage (0 to 25 %, 26 to 50 %, 51 to 75 %, 76 to 100 %). Foliar nitrogen and water content were determined and the resin flow of spruce stems was measured as one indicator of tree defense responses.

The length of the shoots decreased gradually from an average of 9 cm in 2009 (oldest shoots with needles) to 6 cm in 2013 (new shoots). The needle dry mass increased with increasing damage class, while both needle water and nitrogen contents decreased by about 10 % from damage class I to damage class IV. Within damage classes I and II (the prevailing classes), resin flows were higher in trees with increasing loss of needles, indicating that the trees responded to sawfly feeding. However, stem resin flows decreased

in trees with high feeding damage (class III) and were specifically low in trees infested by bark beetles. Chronic defoliation can eventually exhaust plant resources for shoot and leaf production, leading to host decline and death due to additional stress agents (e.g. secondary pests like spruce bark beetles).

The collected data clearly demonstrate a shift in the population dynamics of two sawfly species in lowland secondary spruce forests of Austria and Bavaria. The former dominant *P. abietina* has almost disappeared, while the recent outbreaks are caused by *P. montanus*. The reasons for these unprecedented and synchronous infestations of *P. montanus* in such a large area are unknown and deserve further investigations.

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Detection of Asian Wood Boring Beetles in High-Risk Sites: Two Case Studies from Italy

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ABSTRACT

We presented an overview of a 2-year project (2012-2013) conducted in Italy in collaboration between the University of Padova and the Regional Plant Protection Organizations, aimed at improving the efficiency of the early detection for alien insects. We focused on wood boring beetles (mainly Cerambycidae, Scolytinae and Buprestidae) which include a high number of invasive species (Kirkendall and Faccoli 2010) and represent an important threat to the biosecurity of all forested countries (Brockerhoff et al. 2006). The early detection of alien species is of primary importance to improve the chance of eradication and is usually achieved through direct inspection methods and other sampling strategies such as trapping (Haack 2001, Brockerhoff et al. 2006, Rabaglia et al. 2008, Rassati et al. 2013). Anyway, the resources that can be used for these activities are limited and, at the same time, the number of sites which should be monitored is extremely high. For this reason, it is necessary to find the most vulnerable locations where to concentrate the surveillance efforts.

In 2012, we focused on ports and their surrounding areas, in order to better understand how the port size, in terms of amount of imported commodities, and the characteristics of the landscape surrounding the ports, in terms of forest cover and forest composition, influence the occurrence of alien wood boring beetles. From May to September, 15 Italian international ports were monitored with three traps in each port and three more traps in forest located 3-5 km away from each port. The traps were baited with a generic multi-lure blend proved to be effective in a previous study (Rassati et al. 2012). A total of 81 species were trapped, 67 native and 14 alien. Each forest site close to the port was classified into broadleaf or coniferous forest according to tree composition. The number of alien species was positively influenced by the volume of imported commodities, the position of the trap and the composition of the forest close to the ports. In particular, we found that the mean number of exotic species trapped per trap check was higher in forests than in ports when the forests were mainly composed by broadleaf species, while these values were similar for coniferous forest. Forest cover had no effect. The results suggest that surveillance should be concentrated in large ports and in the surrounding broadleaf forests to improve the cost-effectiveness of the programs.

In 2013, the same experimental scheme applied in 2012 was used to explore the role of wood waste landfills in wood boring beetles' invasion process. In the last 30 years, cargo has increasingly shipped in large containers (Stanaway et al. 2001) and only a fraction of the latter are opened and inspected within ports. Often commodities and associated wood packaging materials are transported directly to their final destinations, such as industrial or commercial areas. Then wood packaging materials are sent to wood waste landfills where they can act as a source of alien wood boring species before being destroyed or recycled. In order to understand the potential impact of the latter sites, a subset of 11 ports from those used in 2012 was selected, and within each site the port and a wood waste landfill were monitored using the same number of traps. We found that the mean number of alien species trapped in ports was similar to that in

wood waste landfills, and that communities did not differ either. This suggests that wood waste landfills have the same importance as ports in wood boring beetles' invasion process, representing an alternative way to detect alien species associated with wood material, even in continental areas far from the coast.

This knowledge may help prioritizing the sites where early detection should take place. These indications have been delivered to the National and Regional Plant Protection Organizations and Quarantine laboratories for further development.

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Nanofabricated Decoys Used in an Electrocution Trap for Emerald Ash Borer and Other *Agrilus* spp.

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ABSTRACT

Here we describe advances in the development of visual decoy based trapping systems for forest buprestid beetle pests, including the emerald ash borer (EAB), *Agrilus planipennis*, an invasive tree-killing pest in North America, and the two-spotted oak borer (TSOB), *Agrilus biguttatus*, a native European oak pest. Like many other buprestid beetles, males of these two species perform a visually-mediated ritualized mating flight directly onto females poised on sunlit leaves. We have successfully evoked this behavior using dead beetles pinned to leaves or green plastic surfaces. Furthermore, when such decoys and the surrounding surfaces are covered by sticky glue, approaching beetles can be trapped (Lelito et al., 2008, Domingue et al., 2013a,b). To provide more user friendly-traps that can be mass-produced, we are now attempting to manufacture synthetic decoys that are capable of attracting male buprestids and to develop glue-free trap designs.

Three types of decoys were developed, the first two by a nano-fabrication process, and the last by using three dimensional (3D) printing. To fabricate the nanofabricated decoys, a dye was created by misting an EAB female beetle with nickel and then removing the biological substrate. For decoys of the first type, a PET sheet coated with a Bragg-stack reflector was loosely stamped over the dye. For decoys of the second type, a green-painted PET sheet was heavily stamped by the same dye. Every decoy of these two types had an underlying black absorber layer. Decoys of the third type were produced by a rapid prototyping machine and 3D printer before being painted green. Fine-scale features were absent on the third type. Experiments were performed in an American ash forest infested with EAB, and a European oak forest with an endemic population of TSOB. When pinned to leaves, dead EAB females, dead TSOB females, and bioreplicated decoys of both types often evoked the complete ritualized flight behavior. Males also initiated approaches to the rapidly prototyped decoy, but would divert elsewhere without making contact. The attraction of the bioreplicated decoys was also demonstrated by providing a high dc voltage across the decoys that stunned and killed approaching beetles. Thus, true bioreplication with fine-scale features is necessary to fully evoke ritualized visual responses in insects, and provides an opportunity for developing insect-trapping technologies.

Sticky branch traps consisting of two 5 X 9 cm² green plastic surfaces affixed roof-like on top of sunlit branches were also deployed using the 3D-printed decoys to target only EAB. Blank traps were compared to identical traps with real EAB females or 3D-printed models added as decoys. Traps of the three varieties were placed on neighboring branches along with one (Z)-3-hexen-1-ol lure per tree. Both real and 3-D printed decoys similarly increased EAB trap captures versus control traps. Daily trap-capture patterns showed sparse activity of EAB before June 18th, followed by a peak in captures of both males and females until June 28th. Beginning at approximately July 1st,

there was a second peak of EAB captures, which consisted almost entirely of males caught on the decoy-baited traps. The native ash borer *Agrilus subcinctus* was found earlier in the season and was also positively attracted to the real and 3D-printed decoys. The results demonstrate the successful application of a small, inexpensive, and fully synthetic decoy-based branch-trap system for EAB.

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Out of the Cities and into the Forest: Recent Detections in Urban Forests of British Columbia

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ABSTRACT

Since 1994, 10 species of introduced Coleoptera, 9 species of Lepidoptera; 4 species of Hymenoptera and one Dipteran have been recorded for the first time in the urban forests of British Columbia. Likely pathways of introduction and detection methodologies are discussed.

Five of the non-indigenous Coleoptera are ambrosia beetles (Curculionidae: Scolytinae) likely introduced in association with wood packaging or dunnage and were detected prior to the implementation of ISPM 15. Four of the five ambrosia beetles are now common in the native forests surrounding greater Vancouver. An additional two species of bark beetles (Scolytinae), *Scolytus schevyrewi* and *Hylastes opacus*, were discovered well after the implementation of compulsory treatment of wood-packaging. While either species may have been introduced on untreated wood-packaging, plausible alternative pathways exist. *Scolytus schevyrewi* may have been transported on untreated firewood from other infested regions of North America while *H. opacus* could have arrived from eastern North America in association with ornamental pines. Two of the remaining three beetle species, the weevils *Phyllobius intrusus* and *Polydrusus sericeus* (Curculionidae: Entiminae), feed on the roots of Cupressaceae and hardwoods as larvae respectively and on foliage of their larval hosts as adults. *Phyllobius intrusus* was first detected in 1994 and is now commonly found feeding on *Thuja plicata* in native forests adjacent to Vancouver. Both Entiminae were most likely introduced in association with horticultural imports. One leaf mining weevil attacking *Ulmus*, *Orchestes alni*, is also present in urban areas in the interior of the province. As importation of elms into BC are restricted to prevent the introduction of Dutch elm disease, the only plausible introduction pathway for *O. alni* is as adventive overwintering adults on other commodities.

Three of the introduced Lepidoptera, *Chrysoclista linneella*, *Macrosaccus robiniella* and *Prays fraxinella* feed on non-native ornamental trees (*Tilia*, *Robinia* and *Fraxinus* respectively). Life histories suggest that both *C. linneella* and *P. fraxinella* could only have been introduced on infested ornamental hosts, the former species overwintering as larvae mining the bark and the latter as early instar larvae mining buds. *M. robiniella* could have been introduced as larvae in leaf mines or as adventive overwintering adults. Two of the six remaining Lepidoptera feed on conifers: *Dichelia histrionana* on *Picea* or *Abies* and *Eupithecia pusillata* on *Juniperus*. The former species overwinters as early instar larvae within needles while *E. pusillata* overwinters as eggs on foliage and each could have easily been introduced on ornamental imports. Three of the introduced Lepidoptera, *Argyresthia pruniella*, *Paraswammerdamia lutarea* and *Gypsonoma aceriana*, feed on native deciduous trees and overwinter either as eggs hidden in bark crevices (*A. pruniella*) or as early instar larvae in cryptic hibernaculae. Each was likely introduced with infested ornamentals. The final non-indigenous Lepidopteran detected, *Psyche casta* is polyphagous on broadleaf trees and shrubs. It overwinters as an early instar larva in a case and could have been transported either as an adventive or in association with ornamental plants. *Psyche casta* has been detected on *Alnus rubra* and *Populus trichocarpa* in native forests well beyond urban areas.

Since 2009, four species of Tenthredinidae, *Monsoma pulveratum* (a defoliator of *Alnus*), *Heterarthrus vagans* (a leafminer on *Alnus*), *Pristiphora geniculata* (a defoliator of *Sorbus*) and *Nematus tibialis* (a defoliator of *Robinia*) have been confirmed to be established in British Columbia. All of these sawflies overwinter as larvae either in a cell in wood (*M. pulveratum*) or as a cocooned larva. The two alder associated species were not likely introduced in association with live plant imports as alders are not valued as ornamentals. As *H. vagans* constructs its overwintering cocoon within the leaf-mine, it is suspected that this species was introduced in leaf debris contaminating some other commodity. Similarly, the overwintering habits of *M. pulveratum* suggests that it was introduced with untreated wood packaging. The remaining two sawflies were most likely introduced in association with horticultural imports as cultivars of both host taxa are widely planted as ornamentals. Each of the sawflies attacking native tree genera has been recovered from native forests well beyond urban boundaries.

A single species of Cecidomyiidae, *Obolodiplosis robiniae*, which causes leaf galls on *Robinia pseudoacacia* is established in British Columbia. *Robinia* is widely planted both as an ornamental in urban areas and as component of reclamation projects around mines and smelters in the province. This gall midge overwinters as a cocooned larva in the soil and was likely introduced in association with live plants. Like *M. robiniella* and *N. tibialis*, it is widespread in southern British Columbia.

Multiple surveillance tools have been used in documenting these introductions. The bark and ambrosia beetles were detected using a combination of trapping and rearing of infested host material. The root weevils were detected through incidental trap captures or beating and *O. alni* was first recovered through rearing of infested plant material. Seven of the introduced Lepidoptera were first recognized through DNA barcoding of light trap captures (6 species) or specimens from reference collections (*E. pusillata*). Only two moth species, *C. linneella* and *M. robiniella*, were recovered during host searches. In contrast, all of the sawflies and the single cecidomyid reported were first detected through host searching and rearing.

Classical Biocontrol of Emerald Ash Borer in North America: Monitoring Demonstrates Successful Establishment

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ABSTRACT

The emerald ash borer (EAB), *Agrilus planipennis* (Coleoptera: Buprestidae), a phloem-feeding beetle native to Asia, is a serious invasive pest of ash trees (*Fraxinus* spp., Lamiales: Oleaceae) in North America. This buprestid was first discovered in 2002 in southeast Michigan and nearby Ontario, where it was determined to be the cause of ash mortality throughout the region. EAB is now known in 22 states and two Canadian provinces where it has killed tens of millions of ash trees. Soon after EAB was discovered in North America, researchers began surveying field populations for insect natural enemies. Although some North American native parasitoids of *Agrilus* spp. attack EAB larvae, prevalence is generally low, and no native egg parasitoids are known (Duan et al. 2009; USDA FS NRS 2014). In recent years, however, species of solitary larval ectoparasitoids in the genus *Atanycolus* (Braconidae), and the solitary larval endoparasitoid *Phasgonophora sulcata* (Chalcididae), have become increasingly abundant in EAB larval populations in southern Lower Michigan (Cappaert and McCullough 2009; Duan et al. 2012a). In the northeast provinces of China where EAB is native, its most prevalent parasitoids are the larval endoparasitoid *Tetrastichus planipennisi* (Eulophidae) and the egg parasitoid *Oobius agrili* (Encyrtidae). A third species, the larval ectoparasitoid *Spathius agrili* (Braconidae), is more common further south in Tianjin where EAB attacks *F. velutina*, a species of ash from the southwestern U.S. and used for landscaping in this region of China (Liu et al. 2003, 2007). Anticipating the need for classical biocontrol as a management tool of EAB in North America, researchers studied the biology, rearing methods, host ranges, and risk assessment of the three parasitoid species. After regulatory requirements were met in July 2007 (Federal Register 2007), APHIS approved their introduction in Michigan (Bauer et al. 2008; Bauer et al. in press).

The three biocontrol agents were released in different EAB-infested ash stands in Michigan's Lower Peninsula during the 2007 field season. Over the next two years, releases expanded to Ohio, Indiana, Illinois, and Maryland, and once establishment was confirmed, USDA initiated the EAB Biological Control Program (USDA FS NRS 2014). This paved the way for APHIS PPQ to launch an EAB Biocontrol Facility in Brighton, MI where the parasitoids are mass-reared for release by cooperators (Fig. 1). Guidelines with information on EAB, parasitoid species, site selection parameters, release and recovery methods, data-entry requirements, and mapping are available online (mapBioControl 2014). As of November 2013, the parasitoids had been released in 18 of the 22 known EAB-infested states and in Ontario, Canada (Fig. 2).

In 2008, we began studying the effects of the biocontrol agents on EAB population dynamics at

study sites in Lower Michigan where releases were done from 2007 to 2010. To estimate egg parasitism, EAB eggs were sampled from the bark of ash trees, and by 2012, we found egg parasitism by *O. agrili* increased from <1% in 2008 to ~19% in 2012 in release plots and ~4% in control plots (unpublished data). To estimate larval parasitism, infested ash trees were felled and debarked, larvae and parasitoids were collected, larvae were dissected, and parasitoids were reared to the adult stage for identification. Larval parasitism by *T. planipennisi* increased from <1% in 2008 to ~21% in 2012 in the release plots, and from 0% in 2008 to ~13% in 2012 in the control in ~13% in control plots (Duan et al. 2013). On the other hand, *Spathius agrili* has not been recovered in ash trees at these field sites, thus it is either undetectable using our sampling methods or not established in Lower Michigan. The most prevalent native insect natural enemies attacking EAB at these study sites were the larval parasitoids *Atanycolus* spp. and *P. sulcata*.

By employing these and other sampling methods after releases were completed, *T. planipennisi* was recovered at other release sites in other areas of Michigan, Ohio, Illinois, Indiana, Maryland, Minnesota, New York, and Wisconsin. Similarly, *O. agrili* was recovered at release sites in Lower and Upper Michigan, Maryland, Ohio, Indiana, Pennsylvania, and New York. APHIS will continue rearing *S. agrili* as an EAB biocontrol agent, however, releases will be restricted to counties on or below the 40th parallel (mapbiocontrol 2014). *Spathius galinae*, a larval parasitoid of EAB from the Russian Far East, is expected to be more cold-hardy than *S. agrili* (Duan et al. 2012b). The Environmental Assessment for release of *S. galinae* in the U.S. is expected to be posted on the Federal Register for public comment in 2014.

Monitoring of established EAB parasitoid populations is essential to the long-term success of the EAB biological control program. It maximizes the efficient use of limited parasitoid resources and provides valuable information to optimize the parameters for successful future releases.

Figure 1. Annual production of *Spathius agrili*, *Tetrastichus planipennisi*, and *Oobius agrili* by APHIS' EAB Biocontrol Facility in Brighton, MI since it first became operational in 2009. Parasitoids are shipped to states for release by researchers and cooperating land managers.

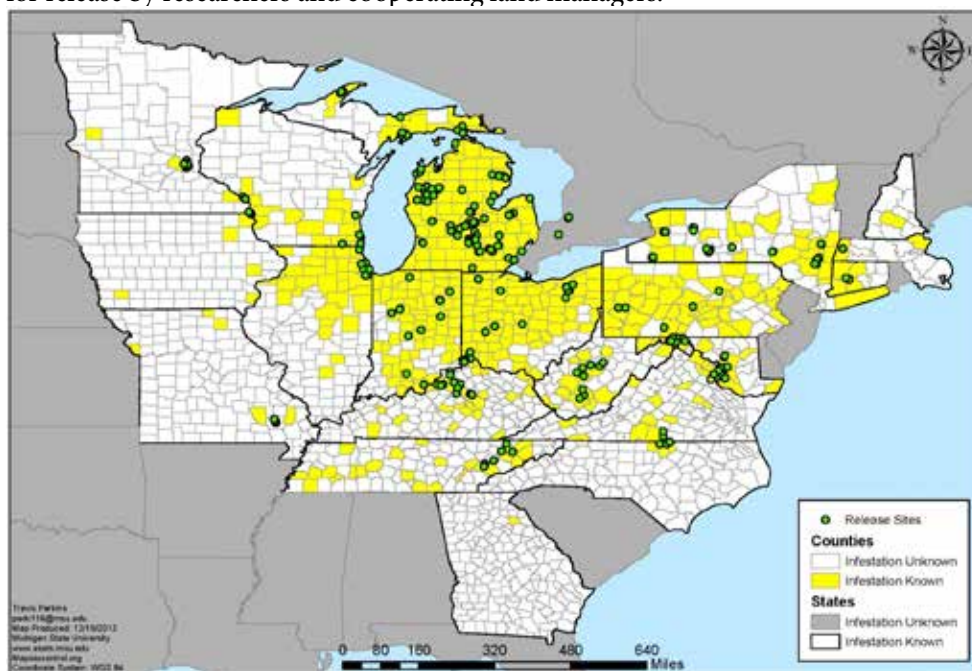


Figure 1. Map of EAB-infested counties in the U.S. and sites where EAB biocontrol agents were released in North America from 2007 to 2013 (Map credit: Michigan State University and USDA APHIS PPQ).

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Unexpected Rapid Appearance and Spread of *Entomophaga maimaiga* in Central-Eastern European Gypsy Moth Populations

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ABSTRACT

The gypsy moth (*Lymantria dispar* L.) is one of the most serious defoliating forest pest in many European countries and was accidentally introduced into the United States in the 2nd half of the 19th century. Its main hosts are the oaks, but it causes damage to many other broadleaved species and sometimes even conifers. Gypsy moth outbreaks are common events both in the species' native and alien ranges and the temporal patterns of outbreaks are similar. Most populations exhibit periodicity with outbreaks recurring every 8 to 12 years. It has also been noted that in the highly susceptible sites (the very dry sites), however, populations exhibit a "doubled" frequency such that outbreaks recur every four to six years. During an outbreak period it may cause severe defoliation on millions of hectares in Central-Eastern Europe. The magnitude of its damage within Europe is greater from west to east and from north to south. The largest outbreak in Hungary (212,000 hectares) was recorded in 2005. The affected area is predicted to increase significantly due to climate change, and based on climate change scenarios, it is also likely that outbreaks will become more frequent and longer in duration. More frequent and lengthy outbreaks will enhance tree mortality and forest decline, causing significant economic and ecological damage. A significant portion of the human population has demonstrated sensitivity to the hairs and scales of gypsy moth life stages and tourism might also be negatively affected by large scale defoliations. These facts suggest that there will be a significant increase in the importance of this species in the future.

Traditional aerial spray control of gypsy moth populations is very expensive (ca. 50-70 Euros/ha) and, more importantly, even the use of "nature-friendly" technologies such as growth regulators or *B.t.* products may have harmful side effects on forest ecosystems. The more selective control technologies (i.e. spraying with NPV, or mating disruption with sex pheromones) which are commonly used in the U.S, are considered as experimental in Europe. Additionally, the EU is considering banning any aerial spraying for plant protection purposes. Therefore, although there is an urgent need for controlling gypsy moth populations now and in the future, there is a lack of sustainable and widely acceptable technologies available to managers for their immediate use.

The entomopathogenic fungus, *Entomophaga maimaiga* was originally described from *L. dispar japonensis* populations collected in central Honshu, Japan, in 1984. This fungus was inten-

tionally introduced into the United States as a classical biological control agent in 1910-1911 however it was never recovered. Beginning in 1989, the fungus suddenly appeared in several states in the Northeast US and since then, it has spread to the south and west and is now found in most of the generally infested gypsy moth region. This fungus has had a major detrimental impact on gypsy moth populations and has caused a major decrease in defoliation throughout this area.

In 1999 (after an apparently unsuccessful attempt in 1996) the fungus was successfully introduced into Bulgaria from the United States into Bulgaria and was followed by several other introductions. Consequently, *E. maimaga* is now common and widespread in Bulgaria and apparently has spread across the borders of Bulgaria in several directions. In 2005 the fungus was unexpectedly recorded from Georgia, and in 2011 was first found in Turkey. An extensive programme was initiated in Serbia in 2011 that involved deliberate introduction at 90 locations. In 2012, *E. maimaga* was first recorded from Greece and Macedonia. In early summer, 2013 natural spread of *E. maimaga* reached Croatia and Slovakia. During this time, it was also recorded at a number of different locations in Hungary.

In all countries listed above *E. maimaga* caused significant (50-100%) mortality in gypsy moth larval populations. As a result, no other microbiological or chemical controls have been necessary in Bulgaria since the introduction of the fungus. The area infested by gypsy moth during the last two outbreaks (after the introduction) is only 15-30% of the average area infested during previous outbreaks. These facts suggest that *E. maimaga* can be a major contributor in the control of gypsy moth populations in Europe, similar to what has occurred in North America.

Entomophaga maimaga is considered to be highly specific to *L. dispar* in the United States. According to the results of extensive studies conducted both in the laboratory and in the field, it very rarely infects any lepidopteran host other than the gypsy moth. Due to their extremely long persistence in the environment, resting spores can be stored for long periods of time and then can be released intentionally at specific locations in order to establish the fungus in local populations and reduce the probability of future outbreaks. However, direct and indirect impacts of *E. maimaga* on non-target and beneficial organisms must be carefully investigated in its projected new area prior to further deliberate releases. Although *E. maimaga* can be considered as having a significant negative impact on its host *Lymantria dispar*, it may also cause basic changes in the community structure of folivore insect guilds on oaks, resulting in increasing populations of other defoliating insect groups such as tortricids, geometrids and sawflies. These potential changes should also be studied. There are several important questions to be answered concerning the distribution and ecological impact of *Entomophaga maimaga* in Europe:

- Mapping the distribution and spread of *Entomophaga maimaga* in Hungary
- Determining the ecological host range of *Entomophaga maimaga*
- Testing physiological host range of *Entomophaga maimaga* in the laboratory
- Testing the impact of *Entomophaga maimaga* on native parasitoids
- Long term changes in community structures of the oak-defoliating guild

Synthesis of Research on *Sirex noctilio*

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ABSTRACT

Sirex noctilio was captured in a trap in Oswego County, New York State in fall 2004 (Hoebeke et al. 2005) and by 2014 it had spread to six more states: Pennsylvania, Vermont, Michigan, Ohio, Connecticut and New Jersey (NAPIS, 2013). *Sirex noctilio* had already been introduced from Europe to countries in the Southern Hemisphere since its introduction to New Zealand around 1900. By now, this invasive has been introduced to 9 countries in both hemispheres. However, this well-established introduction to North America is the first introduction to an area where pines, siricids and their associated communities are native.

Scientists have responded to the potential damage to pine forests that this invasive can cause by conducting research along numerous avenues. In recent years syntheses of this research have been published in printed and digital form. Slippers et al. (2012) edited a book summarizing the basics about *S. noctilio*, its symbiotic fungus *Amylostereum areolatum* and associated parasitic nematodes and hymenopteran parasitoids as well as its occurrence in countries in the Southern Hemisphere. Also in 2012, Schiff et al. completed an online revision of the Siricidae for the Western Hemisphere and this is adding significantly to our understanding of the native North American siricids, which have not previously been studied extensively. In 2013, a symposium on *Sirex* research was held at Cornell University (<http://blogs.cornell.edu/hajek/sirex-symposium/>). In addition, a website on siricids was constructed in 2013 (www.woodwasps.com) to facilitate sharing recent research activities and results. Another website maintained by the South African Forestry and Agricultural Biotechnology Institute (FABI) lists literature on *Sirex*, including both new publications and pdfs of publications that are older and difficult to obtain.

Here, I will summarize some major research developments from my research group as well as some recent research of special interest in North America. With any invasive species, first questions are always "Who introduced it or how did it get introduced?" Molecular studies have now demonstrated that the invasion history of *S. noctilio* is more complicated than expected with independent introductions from Europe that subsequently spread, sometimes resulting in mixtures of genotypes. Boisson et al. (2012) found that two major gene pools were the source of *S. noctilio* introductions and one of these gene pools was from a source population not included in their studies. In the most likely introduction scenario (#5), Boissin et al. (2012) hypothesized that *S. noctilio* was introduced three times from Europe (to North America, South America and Oceania). In this scenario, at least one genotype of *S. noctilio* that is present now in North America probably came directly from Europe and it is possible that the second genotype presently in North America arrived from South America (although it is also a possibility that this second genotype could have come directly from Europe).

It is not always easy to find *S. noctilio* infestations in northeastern North America. To locate infested trees, the obvious resin streaking known to occur at times in the Southern Hemisphere is not normally seen in North America, perhaps due to pine species, lower densities of attacks or the poor health of pines

that are attacked in northeastern North America (D. Haugen, M. Ayres, F. Krivak & K. Dodds, personal communication). It can also be difficult to find *S. noctilio* by collecting adults, which do not fly for a very long period. Intercept panel traps or Lindgren funnel traps baited with ethanol and alpha/beta pinene are generally used to collect adults, but this strategy, which only collects some of the females that are flying, needs improvement. At present, a volatile male-produced pheromone, attractive to both sexes, has been identified (Cooperband et al. 2012) and kairomones from stressed pines are also attractive (Zylstra et al. 2010; Boroczky et al. 2012). Recent wind tunnel studies at Cornell demonstrated a dose response for the male-produced pheromone in two types of traps and adding an ultraviolet light to any trap resulted in enhanced attraction.

For many years, the majority opinion was that each siricid woodwasp was only associated with one species of fungal symbiont (an obligate relationship for the siricid). Recent papers have shown that this is incorrect. Two species of native eastern North American siricids (the pine specialist *Sirex nigricornis*, and the spruce specialist *Sirex nitidus*) were always assumed to be associated exclusively with *Amylostereum chailletii* but we now know that they are at times commonly associated with *A. areolatum*, the fungal species carried by *S. noctilio* (Hajek et al. 2013). There are numerous identifiable strains of these *Amylostereum* species and eastern North American native *Sirex* carrying *A. areolatum* have often been found carrying a newly described native strain. In addition, the pine specialist *S. nigricornis* has also been found carrying one of the strains of *A. areolatum* thought to have been introduced with *S. noctilio*. At present, Louela Castrillo has been investigating the genetic diversity of *A. areolatum* in Europe and comparing it with that in North American genotypes. Her results confirm that there were probably multiple introductions of *A. areolatum* to North America.

The parasitic nematode, *Deladenus siricidicola*, is also the subject of very active research. This nematode has two forms: one that is mycophagous and persists in trees feeding on *A. areolatum* and one that is parasitic on *S. noctilio* when larvae are present. A strain of this nematode from Hungary that sterilizes female *S. noctilio* has been used extensively for biological control in the Southern Hemisphere. While an introduction of this strain (Kamona) was being considered, a non-sterilizing strain of the same species was discovered in North America, probably introduced with *S. noctilio* (Yu et al. 2009). Our investigations at 7 sites in New York and Pennsylvania documented that this non-sterilizing strain was present in 44% of the pines sampled and in 27% of the total *S. noctilio* emerging from these trees (Kroll et al. 2013). The non-sterilizing strain decreases the sizes of *Sirex* females and somewhat reduces the number of eggs produced.

Native species of *Deladenus* are associated with the native *Sirex* in eastern North America, with a different nematode associated with each native *Sirex* species (Morris et al. 2013). Of particular interest, with limited sampling, the non-sterilizing strain of *D. siricidicola* was found, parasitizing *S. nigricornis*, and the native *D. proximus*, which usually parasitizes *S. nigricornis*, was found parasitizing *S. noctilio*. Studies have been conducted of the fitness of *D. siricidicola* and *D. proximus* when feeding on different *Amylostereum* species and strains because the symbiotic fungus present could impact nematode distribution and activity (Morris et al. 2012, 2014). Results suggest that the biological control strain of *D. siricidicola* (Kamona) might not grow well on the strain of *A. areolatum* usually carried by *S. noctilio* in North America. In addition, *D. proximus*, which previously was thought to eat only *A. chailletii*, will, in fact, successfully grow and reproduce when feeding on some strains of *A. areolatum*.

Finally, a long-term study has been conducted in New York and Pennsylvania by David Williams of USDA APHIS. From 2006-2012, he set up studies each year injecting the biological control strain of *D. siricidicola* (Kamona) into pines infested with *S. noctilio* and collecting adult woodwasps emerging from trees the following spring to evaluate whether they were parasitized by Kamona. Molecular methods are presently being used to evaluate whether nematodes in *S. noctilio* were the sterilizing or non-sterilizing strain (or possibly even the native *D. proximus*).

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Invasion Biology of *Phytophthora cinnamomi* in Eastern US Oak Forests: Its Current and Possible Impact Under Projected Climate Change

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ABSTRACT

Phytophthora cinnamomi is among the most notorious plant pathogens with a global distribution. It survives readily in soil but lacks an aerial life cycle or reproduction in fresh water. This Oomycete pathogen is causing ecosystem destructions such as in western Australia. In other parts of the world, beside in agriculture in forest ecosystems it is the primary pathogen or involved in oak decline and mortality in Mexico and Mediterranean Europe, respectively. In eastern US, it has been noticed since the beginning of the 1900's and circumstantial evidence suggested that this pathogen was introduced with early settlers in early 1800's. Beginning of 1900's widespread chestnut mortality was linked to this pathogen. However, after the introduction of Chestnut blight fungus and its devastating impact, this pathogen received little attention and its impact in oak forests was never investigated.

Today it can be commonly found along the eastern oak forests as north as the 40-latitude range. Numerous plants were found to harbor/be infected by this pathogen. Its current range, interestingly, is delineated where the plant hardiness zone 6 ends; no *P. cinnamomi* is to be found in forests above this range despite extensive sampling during the last decade. This data provides strong evidences of how combination of climatic factors inhibited this pathogens spread belowground. Its impact also appears to vary considerably depending on the environmental factors. For example, when investigated in connection with white oak root health, in zone 6, the pathogen caused significant root reductions in particular when trees were stressed by drought events. In the warmer zone 7, at infected sites, trees had more fine roots suggesting a better plant response possibly due to longer growing season. At low elevation sites, such as in southern Ohio, this pathogen appears to be directly involved in oak decline. Despite seemingly doing nothing in some areas, it is causing fine root dieback, and its impact possibly will become more noticeable as climatic abnormalities continue to change the ecosystem balances, unfortunately mostly in favor of this pathogen.

PRESENTATIONS
Mitigating Pathways for Forest Pests

Evaluation of Pest Reduction After Implementation of ISPM 15

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ABSTRACT

Wood packaging material (WPM, e.g., pallets and crating) used in international trade is the likely pathway by which many exotic (nonnative) bark- and wood-infesting insects have become established beyond their native range (Brockhoff et al. 2006, Haack 2006, Haack et al. 2010). The international community responded to the phytosanitary risk posed by untreated WPM in 2002 by approving International Standards for Phytosanitary Measures No. 15 (ISPM 15), which was entitled “Guidelines for Regulating Wood Packaging Material in International Trade” (IPPC 2002). The original goal of ISPM 15 was to “practically eliminate the risk for most quarantine pests and significantly reduce the risk from a number of other pests that may be associated” with WPM (IPPC 2002). The first country to implement ISPM 15 was New Zealand in 2003, followed by Australia in 2004, and the European Union in 2005. The United States implemented ISPM 15 over a 10-month period from September 2005 to July 2006. As of December 2013, at least 78 countries had implemented ISPM 15.

There have been two substantial revisions of ISPM 15 since 2002, and more changes should appear in 2014. The first major revision was in 2006 when the fumigation treatment was lengthened from 16 to 24 hours (IPPC 2006). The next major revision occurred in 2009 when some of the more important changes included the requirement that only debarked wood should be used for WPM, that debarking should precede fumigation, and that the size of any single piece or residual bark should be no more than 50 cm² unless it is less than 1 cm wide (IPPC 2009). The size restrictions on residual bark were based in part on work by Evans (2007) and Haack and Petrice (2009).

The question of how effective ISPM 15 had been in reducing the incidence of live insects in WPM was addressed recently by a National Center for Ecological Analysis and Synthesis (NCEAS) working group entitled “Effects of Trade Policy on Management of Non-Native Forest Pests and Pathogens” (Brockhoff et al. 2011). NCEAS is located at the University of California–Santa Barbara and this particular working group was supported by a grant from The Nature Conservancy. Results of the working group’s evaluation on the effectiveness of ISPM 15 at reducing border incidence in WPM were recently submitted for publication (Haack et al. in review). Below is a brief summary of the major findings. Two other related papers written by members of this working group include Strutt et al. (2013), which analyzed the impact of ISPM 15 on international trade, and Brockhoff et al. (2014), which evaluated how altering propagule pressure through policies such as ISPM 15 could alter establishment rates of borers such as cerambycids and scolytines.

Overall, few studies have been published on the incidence of borers in WPM, and for those studies that have been published the results are often not comparable because some authors recorded infestation levels on a consignment basis (e.g., similar products within a container; Bulman 1992), while others reported infestation rates per individual WPM items such as individual pallets (Haack and Petrice 2009, Zahid et al. 2008). In the pre-ISPM 15 study by Bulman (1992), which was conducted in New Zealand, about 4.3% of the consignments with WPM had infested WPM. By contrast,

in the two post-ISPM 15 studies, which were conducted in Australia and the United States, about 0.1 to 0.5 % of the individual WPM items were infested (Haack and Petrice 2009, Zahid et al. 2008).

The best database our NCEAS working group found to compare pre- vs. post ISPM 15 interception rates was a database maintained by USDA APHIS known as AQIM (Agriculture Quarantine Inspection Monitoring). The incidence of borers in WPM has been recorded in the AQIM program since 2003. The AQIM program involves random sampling of one or more containers each week at more than 40 cooperating ports, with data recorded on a consignment basis.

In our analysis of the AQIM data (which included nearly 35,000 records), we compared WPM infestation rates from 2003-2009 and found a reduction in borer incidence of 36-52% following ISPM15 implementation, depending on the start date and the countries selected for analysis (Haack et al. in review). In general, the infestation rates of WPM fell from about 0.2% to 0.1%.

Because some individuals and groups had expected even larger reductions, we discussed several factors that could have lowered the apparent impact of ISPM 15, such as pest tolerance of the treatments, unintentional noncompliance, fraud, post-treatment pest colonization of WPM, various data issues in AQIM, and a policy anticipatory effect (Haack et al. in review). In addition, it is possible that the AQIM database simply did not start early enough to show the true impact of ISPM 15. For example, if we consider the 4.3% infestation rate recorded by Bulman (1992) to be more representative of the true situation pre-ISPM 15, then the 0.1% infestation rate post-ISPM 15 would represent a 98% drop.

The difficulty our working group had in finding high-quality data from before and after implementation of ISPM 15 restricted our ability to fully evaluate the effectiveness of ISPM 15. One lesson from this effort is the clear need for well-planned sampling programs to be conducted both before and after implementation of major phytosanitary policies so that their impact can be assessed.

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PRESENTATIONS

Urban Forests: The Front Line for Invasive Species

Belowground Causes and Consequences of European Buckthorn Invasions into Remnant Urban Woodlands

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ABSTRACT

Invasions by exotic shrubs are of widespread concern to the management and conservation of forest resources in the eastern United States. These invaders are not only associated with decreased abundance of native plants, but are also hypothesized to alter many belowground properties via to their high quality leaf litter (i.e., many species of invasive shrubs have leaf litter with high nitrogen and/or calcium concentrations). Confirming these belowground effects, however, is difficult due to both the long periods over which these invasions occur and the absence of data on pre-invaded conditions. Furthermore, short-term manipulative experiments may not be able detect belowground changes that accumulate slowly, and simply comparing belowground differences between areas with and without invasive shrubs, as is often done, cannot distinguish between pre-existing and shrub-induced differences.

We developed a repeatable “paired-point” framework to overcome these complications in an investigation into the belowground causes and consequences of invasion by the exotic shrub *Rhamnus cathartica* L. (European buckthorn; hereafter buckthorn) into remnant woodlands of the Chicago region. We located 97 buckthorn-invaded points throughout the woodlands of this region that varied in invasion severity and paired each with a buckthorn-free reference point having no known history of buckthorn invasion. Invasion severity was quantified as an invaded point’s score along the two major axes of a PCoA ordination constructed from measures of buckthorn basal area, percent cover, height, sapling density, and invasion age (3–62 yr) made at each invaded point. Soil parameters hypothesized to promote and/or respond to buckthorn invasion (moisture, pH, total carbon, total nitrogen, C:N ratio, inorganic nitrogen, calcium, leaf-litter mass, and earthworm biomass; hereafter referred to collectively as RVs) were measured at each point. The difference in values for these RVs between each invaded and uninvaded point of all point pairs was calculated (Δ RVs). A relationship (linear or non-linear) between a Δ RV and severity, i.e., slope $\neq 0$, indicates a buckthorn-induced change. An intercept (value of Δ RV where invasion severity = 0) different from zero indicates a pre-existing difference.

Using this paired-point framework revealed novel insights from short-term data regarding the belowground causes and consequences of buckthorn invasion, illustrating the potential utility of this framework for similar investigations for other slow-growing invasive plant species. The patterns this framework reveals can then be used to guide the manipulative, long-term experiments needed to confirm the belowground causes and consequences of these invasions. For example, we found evidence that

the elevated levels typically found in buckthorn-invaded soils for most of the belowground factors that we measured (soil moisture, pH, total carbon, total nitrogen, calcium, and soil NH_4^+ -N) pre-date invasion and are not buckthorn-induced as hypothesized (i.e., intercepts > 0 and slopes $= 0$). Therefore, studying these factors in the context of whether or not they contribute to buckthorn establishment will likely yield more useful insights than will studying them in the context of buckthorn-induced change. Of these factors, we found evidence that soil calcium plays an important, but currently unrecognized, role in determining where buckthorn establishes. Therefore, the role of this nutrient in driving invasions by buckthorn and other forest plant invaders needs further investigation. We also found no evidence of buckthorn altering either soil C:N ratios or earthworm biomass (slope $= 0$), or of the levels of either of these factors being higher in buckthorn-invaded soils (intercept $= 0$), again contradicting hypotheses.

As for buckthorn-induced change, we did find evidence that buckthorn establishes in areas having both greater spring leaf-litter mass and greater rates of leaf-litter decomposition (intercepts > 0) and then proceeds to accelerate leaf-litter decomposition further, and to alter spring levels of soil NO_3^- -N (slopes $\neq 0$). This potential effect of buckthorn on leaf-litter mass agrees with the near absence of a litter layer often observed beneath the canopies of buckthorn and other invasive shrub species. Given that the majority of biodiversity in forested systems occurs in or below this leaf-litter layer, the consequences of shrub-induced leaf-litter loss on forest biodiversity needs further investigation.

In conclusion, the patterns revealed by using this paired-point framework suggest that buckthorn is not as much of a “driver” of belowground change as once hypothesized (although its effect on leaf-litter mass may be a concern for conservation). This insight has important implications for the overall study of the belowground effects of invasive plants. First, it reveals that broad, overarching conclusions regarding the belowground effects of invasive plants cannot be generated from investigations having low replication and/or limited spatial scope, both of which are characteristics of preceding investigations into the belowground consequences of buckthorn. Second, greater consideration of how the belowground differences often observed between areas with and without invasive plants contribute to initial establishment is needed, rather than simply assuming these differences to be invader-induced.

Detecting the Next Worm in the “Big Apple”

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ABSTRACT

Emerald Ash Borer (*Agrilus planipennis*) and Asian Longhorned Beetle (*Anoplophora glabripennis*) are two examples of exotic invasive pests that kill trees and were first introduced into urban areas. Frequently, invasive insects like these are in an area for 5 to 10 years before they are discovered. Given that urban areas are a likely entry point for the next harmful insect or disease that could impact our forests, we need to consider ways to assess tree health without knowing anything about the stressor. In addition, we need methodology that can quantify incipient stress in trees if we are to identify the next threat to our forests when it arrives.

Figure 1 outlines key components of tree health assessment which

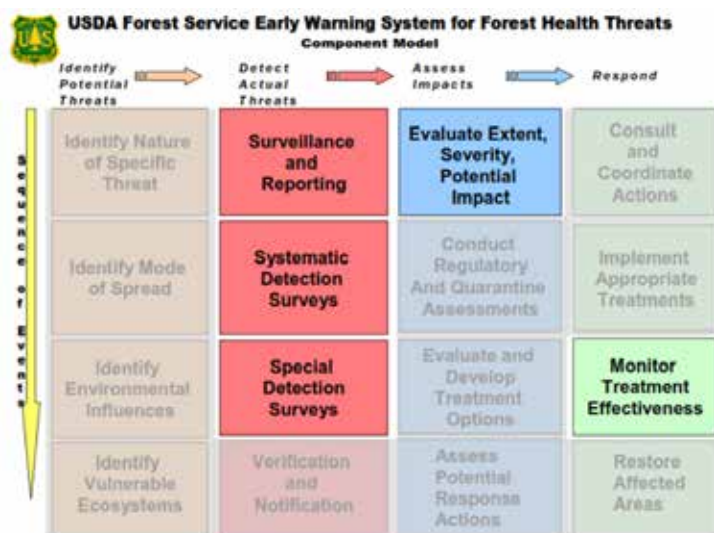


Figure 1. Diagram of the Early Warning System for forest health threats in the United States.

general physiological stress symptoms in a tree. Methods must finely quantify stress symptoms ranging from very early, pre-visual stress all the way through tree mortality. In addition, methods must be flexible and adaptable to different situations and species. This, in turn, will allow for tree health surveys that are broadly applicable across a landscape or cityscape. If these criteria are met we can deploy them in the field at an individual tree or plot level. These data provide information that can be used to calibrate data from remote sensing platforms. It should be pointed out that remote sensing technology shows promise in this area

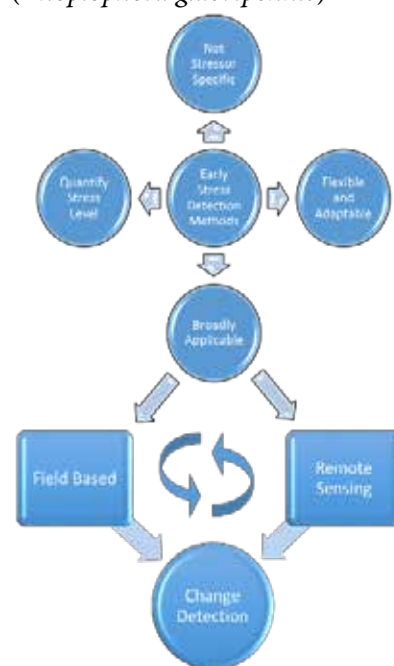


Figure 1. Key components of tree health assessment for earlier pest detection.

need to be in place if we are to detect the next insect or disease threat before it becomes well established. Again, given that we don't necessarily know what the next new stressor will be, the methodology needs to quantify gen-

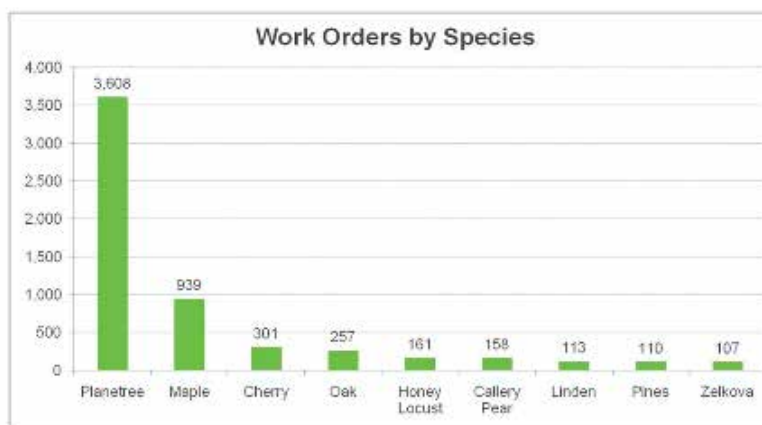
(Pontius et al., 2005, 2008), however, it is not operationally viable yet. If we can meet the criteria outlined in the top part of Figure 1, we can begin to think about surveys of the same areas across multiple growing seasons which will allow for detecting small declines in tree health over time.

In a report by the USDA Forest Service, titled “The Early Warning System for Forest Health Threats in the United States” the authors provided a component model consisting of an ordered series of functional components arranged in loose chronological sequence for early pest detection and response (Figure 2). The component model represents a holistic approach to forest health threats. Figure 1 has been adapted here to highlight only the components that could benefit from finer scale tree health assessment methods.

In order to test the concepts outlined above, we conducted a case study of trees in Queens, NY that were flooded by salt water during Hurricane Sandy. This case study addresses the “Evaluate Extent, Severity, Potential Impact” component in Figure 2. In this case, the components in column 1 “Potential Threats” had been identified or at least hypothesized.

The NYC Department of Parks and Recreation completed a survey of trees that were flooded during Hurricane Sandy and documented that many trees in the flood zone exhibited the symptom of late leaf out (Figure 3). In order to better understand the impacts of salt water inundation, we decided to focus on red maple (*Acer rubrum*) street trees. We used a method that meets the criteria in Figure 2 to quantify stress of an abiotic stressor (salt water inundation). We selected 50 trees that were in the flood zone and 50 trees that were outside the flood zone. Figure 4 shows a map of red maple trees inside and outside the flood zone. We selected our sample trees from each of those areas in Queens, NY.

We measured each of the variables listed in Table 1 for each tree. These variables were com-



Post-Sandy Late Leaf Out Survey

Figure 1. Survey results for trees exhibiting late leaf out symptoms in flood zone.

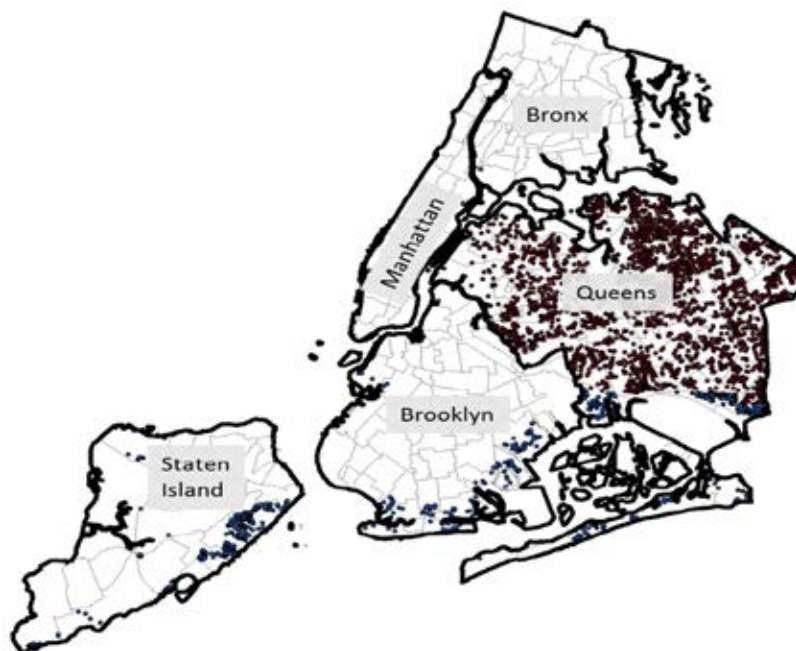


Figure 1. Study area in Queens, NY. Red maple trees in areas that were not flooded (red circles) and areas that were flooded (blue circles).

bined into a single health rating metric (z-score) using the statistical technique described in Pontius and Hallett (2014). We documented a significant difference in tree health between trees that were flooded and that were not flooded (Figure 5).

Each variable contributes to the overall stress rating of the tree. Performance index (PI) and Fv/Fm were measured using a Handy PEA Chlorophyll Fluorescence meter (Hansatech Instruments 2001). These variables provide information about the general physiological health of the tree by quantifying its photosynthetic efficiency. These two measurements can help quantify pre-visual stress symptoms.

Variable	Description
PI	Chlorophyll fluorescence
Fv/Fm	Chlorophyll fluorescence
Transparency	Digital photograph
Fine twig dieback	Visual estimate
Discoloration	Visual estimate
Crown vigor	Visual estimate

Table 1. Variables collected on each tree.

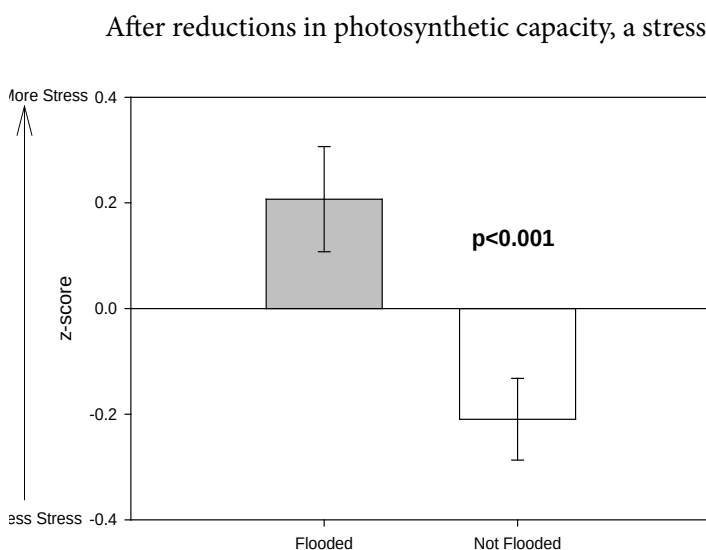


Figure 1. Flooded red maple trees had significantly higher stress than trees that were not flooded.

After reductions in photosynthetic capacity, a stressed tree will begin to reduce foliar biomass. This can be quantified by using digital photographs to quantify the percentage of open versus dark pixels averaged across multiple images of the subject canopy.

As the tree becomes moderately stressed we begin to see an increasing number of fine twigs in the upper and outer portions of the crown without leaves. Fine twig dieback is estimated as a percent of the total crown area.

Foliar discoloration is often a sign of nutrient imbalance, drought stress or in this case a symptom that the flooded trees exhibited. We added this variable to capture the percent of the crown that showed signs of discoloration.



Figure 1. Selected trees with their individual stress ratings. As stress rating increases trees exhibit greater signs of stress.

Finally, we used a visual estimate of overall crown vigor. We used a rubric developed by the North American Maple Project (Cooke et al., 1996). This rates the tree based on several criteria into one of five classes. Class 1 is very healthy and class 5 is dead.

Once the assessment was completed for each tree (5-6 minutes per tree for a team of 2) we standardized the variables into a z-score and summed the z-scores to give each

tree an individual stress rating. Individual tree photos taken at the time of sampling illustrate the progression from healthy to sick as the stress rating increases (Figure 6).

Although the stressor in this case study was not a new pest or disease it did allow us to test our methods against an urban tree health problem. We were able to confirm that trees that were inundated by salt water during Hurricane Sandy were measurably less healthy than individuals that were not flooded. We will continue to rate these trees in subsequent years which will allow us to determine if these trees are on a trajectory of recovery or not. These same techniques can be adapted and deployed in urban settings in order to answer questions about the overall health of our trees. We conclude that this type of survey deployed across a city is one tool that can be used to help with earlier detection of the next invasive threat to our urban trees.

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PRESENTATIONS

Northeast Biocontrol Regional Project (NE-1332):
Updates on Regional Biocontrol Projects

Interactions Between Native Predators and *Cyzenis albicans* in the Control of Winter Moth (*Operophtera brumata*) in the Northeastern US

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ABSTRACT

An outbreaking population of winter moth (*Operophtera brumata*), a non-native geometrid, was identified in eastern Massachusetts in 2004 and since has spread to additional New England states. To control the winter moth outbreak, the Elkinton lab began releases of the biocontrol agent *Cyzenis albicans* beginning in 2005. At one of our 2008 release sites (Wellesely, MA), we have documented a large change in winter moth densities with only a relatively small percent parasitism (35%). Further, in Nova Scotia in the 1950s and British Columbia in the 1970s, the mortality of winter moth due to pupal predation increased following parasitoid establishment. It appears, then, that mortality from parasitism and predation is greater than if the two factors are acting independently; they may be acting synergistically. We tested three hypotheses proposed by Jens Roland as to why this could be: (1) Mortality on winter moth due to parasitoids is enough to bring the population down to a level that can be effectively controlled by generalist ground predators; above the threshold, predators are food-saturated and no longer consume a high proportion of pupae. (2) Parasitized pupae experience lower mortality rates than do the unparasitized pupae. (3) The presence of *C. albicans* induces a numerical response in generalist ground predators because *C. albicans* is a longer-term food source as it overwinters as pupae while winter moth emerges the prior fall. Lastly, we deployed exclusion cages (1/8", 1/4" and 1/2" mesh size as well as open-mesh controls) to estimate which group of predator species may be causing the largest percent mortality.

To test the first hypothesis, we analyzed cumulative pupae mortality at 8 sites in eastern Massachusetts against the corresponding winter moth pupae density. We fit both linear and quadratic polynomial to the data and neither was significant (linear fit: $R^2 = 0.10$, $p = 0.43$, quadratic fit: $R^2 = 0.25$, $p = 0.49$); however, the curvilinear fit hints at a threshold of 125 pupae/m² over which generalist ground predators may be food-saturated. To test the second hypothesis, we compared mortality rates between parasitized and unparasitized pupae. We found that early in the season winter moth was preferentially preyed upon. We currently have *C. albicans* pupae set out for the winter to evaluate over-wintering mortality. To test the third hypothesis, we analyzed the contents of pitfall traps at each site for abundances of ground beetles (*Carabidae*), rove beetles (*Staphylinidae*), ants (*Formicidae*), and shrews (*Blarina brevicauda* and *Sorex cinereus*). With one year of data, there was not significant evidence to suggest that invertebrate predators or small mammal numbers corresponded with winter moth pupae densities across sites (all p -values > 0.05). However, the site with the highest percent *C. albicans* tended to also have high predator abundances. Considering pupae mortality against predator abundance for each site, ground beetle and ant abundances corresponded with higher pupal mortality (p -values < 0.01). The exclusion study controls (open cages) were most heavily predated and there were inconsistent differences between the smaller mesh sizes suggesting that, at the end of the summer, that shrews are causing the largest source of pupal mortality. In conclusion, we have limited support for all three hypotheses. The work shows that it is necessary to account for other sources of mortality when assessing the impact of an introduced biocontrol agent.

Classical Biocontrol of Emerald Ash Borer: A Life Table Analysis of Population Impacts of Introduced Natural Enemies

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ABSTRACT

Life tables are widely used to study the effects of various biotic and abiotic factors in the population dynamics of insect pest populations (e.g., Varley et al. 1973, Southwood and Henderson 2000). By following a cohort of insects throughout its entire life cycle in its natural environment and recording survivorship and/or specific mortalities at each stage, a stage-specific life table can then be constructed. Life tables are then used to elucidate the role that each mortality factor plays in suppressing pest population growth (Hassell 1985, Parker 2000). This type of life table study is particularly useful when implementing biological control programs against invasive insects because such programs require critical information on the effectiveness of each biocontrol agent against the target pest populations (Nielsen et al. 2008, Duan et al. 2010).

The emerald ash borer (EAB), *Agrilus planipennis* Fairmaire, is a serious invasive forest pest that has killed tens of millions of ash (*Fraxinus*) trees in the United States and Canada. In the present study, we established experimental cohorts by caging gravid female EAB on the trunks of healthy green ash trees (*F. pennsylvanica*). We then observed survivorship and mortality of those larvae, along with the fates of naturally occurring (wild) EAB larvae in the same trees. The biotic mortality factors we observed included predation, parasitism, putative host-tree resistance, disease, and intraspecific competition. Our study was conducted in six forested areas of central Michigan, where one egg parasitoid (*Oobius agrili* Zhang and Huang [Hymen: Encyrtidae]) and two larval parasitoids (*Tetrastichus planipennisi* Yang [Hymen.: Eulophidae] and *Spathius agrili* Yang [Hymen.: Braconidae]) had been released as part of a biological control program against EAB (Duan et al. 2010, 2012, 2013). Using separate estimates of adult fecundity and egg mortality, we constructed life tables of both experimentally established cohorts and wild EAB populations and compared the net population growth rates across different study sites. Results of our study showed that the net population growth rate ($R_0 \pm SE$) for the experimental cohorts (16.0 ± 2.9) and associated wild EAB stages (19.4 ± 1.9) were the highest for the first (2008-2009) study period at three Ingham Co. sites, but decreased to 4.7 ± 0.9 and 4.6 ± 0.4 , respectively, for the second (2009-2010) study period at the same sites. This sudden decrease in R_0 of both experimental and wild EAB cohorts in those sites corresponded with increases in parasitism by native parasitoids (*Atanycolus* spp.) (Hymen.: Braconidae) and the introduced biocontrol agent *T. planipennisi*. In contrast, R_0 values of both experimental cohorts (5.7 ± 2.2) and associated wild EAB populations (11.3 ± 2.5) were intermediate in the third (2010 – 2011) generation at different sites in Gratiot and Shiawassee Cos. There was no significant difference in R_0 values between the experimental cohorts and wild EAB populations for the first and second generations. However, R_0 of the experimental cohorts was significantly lower than that of associated wild EAB population for the 3rd generation. Relevance of our findings to EAB population dynamics in the invasion process was discussed.

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PRESENTATIONS

Recent Advances in Quarantine Monitoring and
Management of Exotic Wood Boring Insects

Field Testing of Generic Pheromone Lures for Cerambycids in China

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ABSTRACT

From 2010-2013, we tested the attraction of cerambycid beetles to a library of known cerambycid pheromones in southern China. These multi-year experiments were conducted in two locations: 1) a tropical rain forest in the Bulong Nature Preserve, Yunnan Province, China near the Myanmar/China border, and 2) an urban forest park in the city of Nanning, Guangxi Autonomous Region, China. From 28 May to 25 June 2010, in Yunnan Province, a total of 1,526 cerambycids representing 71 species were captured. Fourteen species were significantly attracted to at least one compound, and accounted for 92% of the specimens collected (Wickham et al., 2014). Compounds with a 3-hydroxyalkan-2-one or 2,3-alkane-diol motif attracted significant numbers of both sexes for eight species in the subfamily Cerambycinae, including species in the genera *Demonax*, *Rhaphuma*, and *Xylotrechus*. Only one species, *Rhaphuma horsfieldi* (White) was strongly attracted to more than one test compound [(2*R**,3*R**)-2,3-hexanediol and (2*R**,3*R**)-2,3-octanediol]. Within the Lamiinae, males and females of five species, including *Acalolepta formosana* (Breuning), *Monochamus bimaculatus* Gahan, *Pharsalia subgemmata* (Thomson), *Pseudomac-rochenus antennatus* (Gahan), and *Xenohammus bimaculatus* Schwarzer, were significantly attracted to 2-(undecyloxy)ethanol (monochamol). This is the first report of significant attraction of species in genera other than *Monochamus* to 2-(undecyloxy)ethanol, suggesting that this pheromone component is more widespread in the subfamily Lamiinae than previously thought. In particular, in extensive field tests of monochamol in the US and Canada over the past several years, only *Monochamus* spp. had been attracted (Allison et al., 2012; Fierke et al. 2012; Macias-Samano et al., 2012). Unexpectedly, only male *Megopis costipennis* White, subfamily Prioninae, were significantly attracted to (2*R**,3*S**)-2,3-octanediol, suggesting that this compound may be a female-produced sex pheromone component for this species. To date, 2,3-octanediols have only been reported as aggregation pheromone components for cerambycine species, although the homologous 2,3-hexanediols are known female-produced sex pheromones or sex attractants for several species in the prionine genus *Tragosoma* (Ray et al. 2012). In 2013, a six component lure [including (2*R**,3*R**)-2,3-hexanediol, (2*R**,3*S**)-2,3-hexanediol, (2*R**,3*R**)-2,3-octanediol, (2*R**,3*S**)-2,3-octanediol, 3-hydroxyalkan-2-one, and monochamol] was tested for 7 days at 22 randomly selected sites over a large (40 km diameter) geographic area, including the nature preserve. A total of 4,577 specimens representing 62 cerambycid species were captured. The traps baited with multicomponent generic lures recaptured a majority of the species attracted in significant numbers to traps baited with single compounds in 2010, including five laminine species, five cerambycine species, and *Megopis costipennis* (the only prionine species). Three of the eight cerambycine taxa captured in significant numbers in 2010 were absent from the 2013 study, includ-

ing the cerambycines *Demonax literatus literatus*, *Demonax ornatus*, and *Xylotrechus atronotatus draconiceps*. These results suggest possible inhibition or antagonism by one or more compounds in the multicomponent lures for a minority of species in the subfamily Cerambycinae, but not for species in the subfamilies Lamiinae or Prioninae. In other words, the presence of multiple diol or hydroxyketone pheromone components may have affected the attraction of some cerambycines, but the same compounds did not inhibit the attraction of lamiines to 2-(undecyloxy)-ethanol, nor attraction of *Megopis costipennis* to racemic (2*R**,3*S**)-2,3-octanediol.

In parallel studies in Guangxi Autonomous Region, China, traps baited with monochamol attracted significant numbers of adult male and female *Monochamus alternatus*, and traps baited with a dialkyl ether structural motif attracted significant numbers of male *Anoplophora chinensis*. Whereas captures of the former species were expected because we had previously identified monochamol as its pheromone (Teale et al., 2011), the pheromone of the latter, very economically important species has not yet been identified. This species is being targeted for full pheromone identification this year, based on this strong lead.

Our results support the hypothesis that pheromone structures and patterns of use are highly conserved within cerambycid taxa, with specific compounds being used as pheromones within a genus, but also among different genera, tribes, and even subfamilies. Furthermore, tests of generic blends of pheromones revealed that the presence of multiple pheromone components may be antagonistic to a few species. Our results also suggest that traps baited with even a limited number of pheromones may be useful for surveying forest biodiversity, as well as for detecting and monitoring specific species, particularly those that have the potential to be invasive pests.

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Early Detection of Cerambycidae and Curculionidae Species in Survey Traps: What are We Missing?

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ABSTRACT

Early detection of invasive species is critical for eradication or effective containment. Here we report results of two trapping experiments testing the effects of trap placement and lure component on the diversity and abundance of bark- and wood-boring beetles (Cerambycidae and Curculionidae) detected in traps. We were particularly interested in comparing treatments for the number and proportion of species detected per trapping effort at each site, i.e., species accumulation curves. Experiment 1 was a factorial experiment testing the effect of ethanol (E), *E/Z*-fusicumol (EZF), *E/Z*-fusicumol acetate (EZFA), and all binary and ternary combinations (8 lure treatments), on species detection. There were eight replicates per treatment and all traps were placed high in the tree canopy of a mixed coniferous-broadleaf forest near Gagetown, NB, Canada, 22 May–24 September 2013. Experiment 2 was a 2 x 2 factorial experiment testing the effects of trap height and lure, replicated six times in a white pine-dominated stand near Mt. Currie, NB, 24 June–17 September 2013. Traps were placed either 1.5 m above the ground or high in the tree canopy and baited with either α -pinene + ethanol (aPET) or a pheromone-enhanced combination of racemic 3-hydroxyhexan-2-one + racemic 3-hydroxyoctan-2-one + racemic *R***R**-2,3-hexane diols + fusicumol + monochamol + ipsenol + aPET. We used 12-funnel Lindgren traps coated with Fluon® with saturated salt solution in the cups in both experiments. We detected 81 species in experiment 1 (46 Cerambycidae and 35 Curculionidae in 64 traps) and 74 species in experiment 2 (39 Cerambycidae and 35 Curculionidae in 24 traps). Species composition was significantly affected by lure, trap height, and lure*trap height interactions. Traps baited with pheromones plus host volatiles (e.g., EZF+E) detected more species per trap than did traps baited with host volatiles or pheromones alone. The number of species detected per trapping effort was improved by using pheromone-enhanced lures and by placing traps both high in the canopy and near the ground. Species accumulation curves rarely reached an asymptote for any treatment even with 8 traps per site, suggesting that surveys that use relatively few traps per site may fail to detect all species that are present, e.g., 3–6 traps per site detected only 35–60% of the total species collected at each site. Studies planned for 2014 will compare the relative costs of high traps vs. low traps.

Enhancing Identification of Cerambycids and Buprestids from Port Interceptions in Solid Wood Packing Material

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ABSTRACT

Despite ISPM-15 regulations requiring treatment of solid wood packing material (WPM) to prevent importation of wood pests, live larvae of potentially destructive exotic wood borers are intercepted at U.S. ports of entry in wooden pallets, crates, and dunnage. Actionable wood borer larvae found during port inspection activities often remain unidentified beyond family level because immature stages are difficult to identify using morphological characteristics, and DNA barcodes are not available for many species. As a result, valuable information on risk pathways and failure of phytosanitary treatments is lost. Through an ongoing cooperative effort between U.S. Customs and Border Protection (CBP) and USDA Animal and Plant Health Inspection Service (APHIS), live larvae in the families Cerambycidae and Buprestidae intercepted at six U.S. ports were sent to the APHIS Center for Plant Health Science and Technology containment facility at Otis Laboratory for rearing to the adult stage. Adult specimens were then sent to the Systematic Entomology Laboratory for morphological identification, and DNA barcodes were developed at Otis Laboratory. Once barcode sequences from identified species are submitted to a database (e.g. GenBank), any life stage of that species can be identified by matching barcode sequences with existing records.

As of December 2013, Otis Laboratory received 336 cerambycid and 36 buprestid larvae from 19 countries and reared 72 adult cerambycids. To date, no buprestid larvae have been reared to the adult stage, suggesting delayed development or inadequate larval collection and/or rearing methods for this group. Cerambycids identified by morphological and/or molecular methods represented 14 genera and 19 species. We also extracted DNA from larvae that died during rearing, identifying 20 additional specimens to species, and 30 to genus level by matching their barcode sequences to existing sequences in databases. DNA sequences from 25% of cerambycid larvae could not be matched with DNA sequences in databases, indicating that adult specimens will be needed to develop taxonomically identifiable barcodes for some larvae. Notable interceptions of cerambycids included serious invasive pests such as Asian longhorned beetle, *Anoplophora glabripennis* (5 interceptions), and velvet longhorned beetle, *Trichoferus campestris* (6 interceptions); other species with pest potential included *Chlorophorus diadema* (subfamily Cerambycinae) and *Monochamus galloprovincialis* (subfamily Lamiinae). A larger number of live cerambycid larvae intercepted in heat-treated wood than in methyl bromide treated wood. However, the majority of larvae intercepted in wood stamped as heat-treated die during rearing, while many larvae intercepted in wood marked as methyl bromide-treated survive to the adult stage. The differences between treatments in proportions of beetles surviving to adulthood suggest residual lethal effects from heat treatment, but not from treatment with methyl bromide. Notably, all ISPM-15 treatment failures for Asian longhorned beetle were traced to a single WPM treatment facility in China, marked as treated with methyl bromide. Other treatment failures are evident, manifesting as multiple interceptions of potentially invasive species in WPM originating from single facilities. This ongoing project is enhancing capacity to identify all life stages, especially of cerambycids, through DNA barcoding, and is helping identify additional species at risk of introduction and discern treatment failures and pathways of introduction. Together these results promote more targeted inspection of WPM and improved compliance with ISPM-15 requirements.

Cerambycid Interceptions and Their Impact in the Netherlands

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ABSTRACT

The Netherlands has adopted the International Standard for Phytosanitary Measures (ISPM) No. 15 for Wood Packing Materials (WPM). This measure reflects the necessity of a heat- or a chemical (Methyl Bromide) treatment of wood thicker than 6 mm and is approved by the International Plant Protection Convention (IPPC). Prior to use WPM should be properly treated and debarked to prevent the spread of live plant pests and diseases, especially Quarantine species. WPM treated according to ISPM 15 is marked, but these measures are not always properly implemented. Interceptions by the plant protection service of the Netherlands of living insects, such as larvae and adults of Cerambycidae, occurs regularly in WPM during import inspections at airports, at harbours or at stone-importing companies. Living larvae and adults of Cerambycidae have been intercepted in wood from numerous tree species, both coniferous and deciduous trees, in particular those used for the transport of stone and tiles from China. In addition to wood packaging, handicrafts manufactured from wood are also a pathway for live Cerambycid larvae.

In this presentation the identifications are discussed of the various cerambycid larvae and adults (Cerambycinae, Lamiinae, Prioninae, Spondyliinae). Often it is not possible to complete the identification of larvae of Cerambycidae, because little diagnostic information is published or available online. Molecular information (PCR, CO1-sequences) is becoming more available and offers an additional possibility to support identifications.

Cerambycids of Quarantine Concern for EPPO Region

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ABSTRACT

EPPO maintains several lists of pests: the Alert List, A1 and A2 Lists of pests recommended for regulation to EPPO countries (A1 for species not present in the EPPO region and A2 for species present in the EPPO region but not widely distributed), and the List of invasive alien plants. All these lists are available on the EPPO website (www.eppo.int). The purpose of the EPPO Alert List is early warning of EPPO countries about pests that could present risk to their territories. It is maintained by the EPPO Secretariat and critically reviewed every year by the EPPO Working Party for Phytosanitary Regulations (when alert has been given and no further action taken, pests are deleted after 3 years on the list). The Working Party also decides about priorities for Pest Risk Analysis (PRA) for the pests of the Alert List. According to the PRA results, a pest is moved to A1 or A2 list or deleted from the Alert List.

In the beginning of 2014, only one cerambycid species is included in the EPPO Alert List:

- *Aromia bungii* (added to the Alert List in 2012), the pest of *Prunus* (preferred), *Azadirachta*, *Bambusa*, *Diospiros*, *Olea*, *Populus*, *Pterocarya*, *Punica* and *Schima*. It is present in China, Korea, Mongolia, Taiwan, Vietnam, and is under eradication in Germany and Italy, and intercepted in UK.

In the beginning of 2014, following cerambycid species are included in the EPPO A1 List:

- *Anoplophora glabripennis* (added to the A1 List in 1999). *Acer*, *Aesculus*, *Betula*, *Populus*, *Salix* and *Ulmus* are preferred hosts, many other trees are damaged. It is present in China, Japan, Korea, Taiwan, USA, and is under eradication in European countries.
- *Apriona cinerea*, *A. germari*, *A. japonica* (added to the A1 List in 2013). *Malus*, *Morus*, *Populus* and *Salix* are preferred, many other trees are damaged. It is present in South-East Asia (*A. japonica* – only Japan, *A. cinerea* – only India and Pakistan).
- *Monochamus* spp. are included as potential vectors of *Bursaphelenchus xylophilus*. Most are present in Canada and the USA (*M. alternatus* and *M. nitens* – in South-East Asia).
- *Oemona hirta* (added to the A1 List in 2010), the pest of over 200 species of trees and shrubs from 81 families, *Citrus* sp. are preferred. It is originally present in New Zealand, and intercepted in UK in *Wisteria* plants.
- *Saperda candida* (added to the A1 List in 2010), the pest of *Malus* (preferred), *Amelanchier*, *Aronia*, *Cotoneaster*, *Crataegus*, *Cydonia*, *Prunus*, *Pyrus*, *Sorbus*. It is present in Canada and the USA, and is under eradication in Germany.

In the beginning of 2014, following cerambycid species are included in the EPPO A2 List:

- *Aeolesthes sarta* (added to the A2 List in 2002). *Juglans*, *Malus*, *Platanus*, *Populus*, *Salix* and *Ulmus* are preferred, many other trees are damaged. It is present in Afghanistan, India, Iran, Kyrgyzstan, Pakistan, Tajikistan, Turkmenistan and Uzbekistan.

- *Anoplophora chinensis* (added to the A2 List in 2007). *Acer*, *Citrus*, *Fagus*, *Malus*, *Populus*, *Salix* and *Ulmus* are preferred hosts, many other trees are damaged. It is present in China, Indonesia, Japan, Korea, Malaysia, Myanmar, Taiwan and Vietnam, established in Italy, and under eradication in some European countries.
- *Hesperophanes campestris* (added to the A2 List in 2007). *Malus* & *Morus* are preferred, *Betula*, *Broussonetia*, *Gleditsia*, *Salix*, *Sorbus* & many other trees damaged. It is present in Armenia, Japan, Kazakhstan, Korea, Kyrgyzstan, Mongolia, Russia (Far East), Tajikistan and Uzbekistan.
- *Tetropium gracilicorne* (added to the A2 List in 2002), the pest of *Abies*, *Larix*, *Picea* and *Pinus*. It is present in China, Japan, Kazakhstan, Korea, Mongolia and Asian Russia.
- *Xylotrechus altaicus* (added to the A2 List in 2002), the pest of many species of *Larix*. It is present in Mongolia and Asian Russia.
- *Xylotrechus namanganensis* (added to the A2 List in 2004). *Alnus*, *Juglans*, *Malus*, *Morus*, *Platanus*, *Populus*, *Prunus*, *Salix*, *Ulmus* are preferred and many other trees damaged. It is present in Afghanistan, China, Iran, Kazakhstan, Kyrgyzstan, Tajikistan, Turkmenistan and Uzbekistan.

Barcoding the Cerambycidae: Progress in the Molecular Identification of Long-horned Woodborers

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ABSTRACT

There are an estimated 1,100 species of long-horned woodborers in North America (NA) and more than 35,000 species globally. While the North American fauna is relatively well known, invasive Cerambycidae such as *Anoplophora glabripennis*, *Callidiellum rufipenne*, *Tetropium fuscum* and *Trichoferus campestris* continue to be discovered in the urban and forest habitats of NA. Although the adults are relatively well known, the immature stages are poorly known and are a challenge to fully identify when intercepted in wood-packaging or dunnage of unknown provenance. Molecular diagnostics provide a means of identifying immature life stages, provided that DNA sequence libraries derived from reliably identified adult voucher specimens are available for comparison.

The data presented for the current status of DNA reference libraries for Cytochrome C oxidase sub-unit I (COI) sequences from reliably identified adult voucher specimens is derived from two projects: a collaboration with the Barcode of Life Network [<http://www.boldsystems.org>] to generate COI sequences from museum specimens in Canadian Forest Service reference collections; and ongoing work with the Quarantine and Invasive Species project generating COI sequences from the Canadian National Collection (Ottawa) led by Dr. P. Bouchard. Sequence data from both projects have been deposited in the Barcode of Life Database (BoLD). The former project sampled 1560 specimens from 246 species and generated 1170 COI barcode sequences (>500 bp in length) for 218 of the species sampled. The latter project sampled 275 species and generated 381 COI barcode sequences from 198 species. When combined with other data sources in BoLD, there are (as of December 2013) 6,830 COI barcode sequences from 1,423 species of Cerambycidae in the BoLD database; only 4% of the global fauna is represented.

Continued development of COI sequence reference libraries for molecular diagnostics will require access to reliably identified adults specimens. As many collections do not allow mining of specimens for generation of sequence data we are appealing to any researchers working with Cerambycidae to provide adult material suitable for DNA extraction to support this ongoing work. Please contact the author at lhumble@nrcan-rncan.gc.ca or Dr. Patrice Bouchard (bouchardpb@agr.gc.ca) if you can supply material for this project.

POSTERS

Detection of Alien Wood Boring Beetles in High-Risk Sites: Two Case Studies from Italy

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ABSTRACT

We presented an overview of a 2-year long project (2012-2013) conducted in Italy in collaboration between the University of Padova and the Regional Plant Protection Organizations, aimed at improving the efficiency of the early detection of alien insects. We focused on wood boring beetles (Cerambycidae, Scolytinae and Buprestidae) which include a high number of invasive species (Kirkendall and Faccoli 2010) and represent an important threat to all forested countries (Brockerhoff et al. 2006). Early detection of alien species is of primary importance to improve the chance of eradication and is usually achieved through direct inspection methods and sampling strategies such as trapping (Haack 2001, Brockerhoff et al. 2006, Rabaglia et al. 2008, Rassati et al. 2013). Resources available for these activities are however limited and the number of sites which should be monitored is extremely high. For this reason, it is necessary to find the most vulnerable locations where to concentrate the surveillance efforts.

In 2012, we focused on ports and surrounding areas, in order to explore how port size, in terms of amount of imported commodities, and characteristics of the landscape surrounding the ports, in terms of forest cover and forest composition, may influence the occurrence of alien wood boring beetles. From May to September, 15 Italian international ports were monitored with multi-funnel black traps, three in each port and three in forests located 3-5 km around each port. Forests were classified into broadleaf or coniferous forest according to tree composition. Traps were baited with a generic multi-lure blend previously tested in a pilot experiment (Rassati et al. 2013). A total of 81 wood boring beetle species were trapped, 67 native and 14 alien. The number of alien species was positively affected by volume of imported commodities, trap position and forest composition. In particular, the mean number of alien species per trap check was higher in broadleaf forests than in ports, but it had similar values in coniferous forest. Forest cover had no effect. The results suggest that surveillance should be concentrated in large ports and in the surrounding broadleaf forests.

In 2013, we explored the role of wood waste landfills in wood boring beetles' invasion process. In the last 30 years, cargo has increasingly shipped in large containers (Stanaway et al. 2001) and only a fraction of the latter are opened and inspected within ports. Commodities and associated wood packaging materials are often transported directly to their final destinations, such as industrial or commercial areas. Wood packaging materials are then sent to wood waste landfills where, before being destroyed or recycled, they can act as a source of alien wood boring species. A subset of 11 ports monitored in 2012 was selected. Each port and the corresponding wood waste landfill were monitored using the same traps and lures than 2012. Number and composition of trapped alien species were similar in ports and in wood waste landfills. This suggests that wood waste landfills have the same importance as ports in wood boring beetles' invasion process, representing a crucial site for detecting alien species associated with wood material, even in continental areas far from the coasts.

This knowledge may help prioritizing the sites where early detection should take place. These indications have been delivered to the National and Regional Plant Protection Organization and Quarantine laboratories for further development.

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Optimizing Resource Allocation by Integrating Pathway Analyses and Economic Impacts

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ABSTRACT

Invasive species introductions and impacts are driven by complex interactions between anthropogenic and biological systems. These interactions pose significant challenges for management and resource allocation. We use an array of models – pathway, habitat, spread, and economic – that provide a robust description of the invasions process to quantify subsequent impacts. The output of each model identifies high risk areas and influences how resources should be allocated along the safeguarding continuum. Pathway analyses determine the level of risk for processes that could result in the introduction of invasive species. Introduction alone, however, is not enough to warrant the allocation of resources. The ability of the pest to find a suitable habitat and establish a population is another critical factor. Once established, the rate of spread and abundance of the pest results in varying level of economic impact. We have compiled data quantifying the pathways for pest introductions; created habitat suitability models for pests and have economic models that project impacts at varying rates of spread. We plan on integrating these models to create a mitigation and decision support tool that will help optimize resource allocation along the safeguarding continuum and to high risk areas.

Drought as a Predisposing Factor for Bark Beetle Attack

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ABSTRACT

Drought periods induce a continuously increasing water stress in trees which - under Central European conditions –become more and more susceptible as hosts for aggressive bark beetle species, like the spruce bark beetle *Ips typographus* (Christiansen and Bakke, 1997). As it is known from various conifers, this indirect effect is due to a lower tree defense reaction resulting from reduced photosynthesis, lower oleoresin monoterpene content (Levinsohn et al., 1991) and a decrease in resin flow (Reeve et al., 1995). The aim of this interdisciplinary project is to find thresholds for the interaction between soil and tree water balance and the susceptibility of the host tree for bark beetle attack which can be used to quantify the actual risks of drought as a predisposing factor for bark beetle attack. At six experimental plots (size: 18x20 m each) in an upper, moderately north-west exposed, 90-year old Norway spruce stand mixed with European beech, a roof construction was used to establish a gradient of water deficit. Therefore, two of these plots were totally covered, two plots were partially covered, and two plots were uncovered and serve as controls. From each plot, three spruce trees were selected for tree physiological analyses and induced attack experiments with bark beetles. In multivariate data analysis we test the hypothesis that a relationship exists between increased soil water deficit, reduced tree defenses and an increased susceptibility of the tree for bark beetle infestation. Combined with the hydrological „Coup Model“, which is based on the climate data, forest stand and soil-related parameters of the study site (Jansson and Karlberg, 2010), the results will allow us to estimate the predisposing effect of drought for bark beetle attack. Furthermore, the field data will be combined with modelled data of the phenology model „PHENIPS“ (model for estimating the development of *Ips typographus*, Baier et al., 2007) and the Predisposition Assessment System „PAS“ (Netherer and Nopp-Mayr, 2005) in order to obtain a dynamic model for risk assessment of site and stand-related conditions for bark beetle outbreaks.

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***Sirex noctilio* (Hymenoptera: Siricidae) Mating Behavior**

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ABSTRACT

During the summers of 2012 and 2013, bioassays were conducted with the invasive pine pest, *Sirex noctilio* (Hymenoptera: Siricidae) to understand mating behavior and the conditions necessary for mating. It has been suggested that light intensity and temperature influence mating behavior of *S. noctilio*. Based on our laboratory observations and previous indoor trials, we hypothesized that the age of the wasps and the ratio between male and female sizes could also influence mating occurrence.

To conduct mating studies, fabric-netting cages (60x60x60 cm) were placed in the shade outdoors with ten males and one female per trial. Each of the 68 trials were each conducted for 16 min, and during this period behavioral data was collected for all wasps every 4 minutes. Temperature and light conditions were also recorded.

Bioassays were conducted on days with differing weather conditions. On days that were partially cloudy and on sunny days the wasps tended to be more active and crowd in the brightest corner of the cage. Higher percentages of females mated under these conditions than on cloudy days. Temperature also affected mating success. Wasps were not very active at lower temperatures and they did not crowd together and fewer females were mated. At temperatures between 31°C and 33°C mating success was higher than 90%. At temperatures higher than 33°C there was a trend for decreasing mating success.

The sizes of males and females of each successful mating event were compared to determine the optimal size ratios (males:females) and 66% of the males that successfully mated were smaller or the same size as the female they mated with.

None of the 1 day old males mated and only a few 2 day old males mated, but the percentage of mating success increased when males were 3-5 days old.

This experiment suggests that in nature *S. noctilio* would mate more often and successfully on sunny and warm days and that it would be less common for small females to mate with larger males. However, another interesting observation made during these trials is that females of this woodwasp seem to need a substrate to hold onto while they are mating. Our observations suggest that they do not mate during flight as some literature has suggested

Containing Domestic Spread of Pests: How Overcome the Challenges?

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ABSTRACT

Once introduced to North America, forest pests spread to additional vulnerable areas via X major pathways: Currently, few of the pests are targeted by control measures, most domestic pathways are not adequately regulated, and policy-makers expect rapid success. Changes are needed to ensure that all serious pests are targeted, that “clean-up” measures are applied to all pathways, and that efforts are started earlier in the invasion process and maintained until solutions are found. All these improvements must be made in a period of shrinking resources for federal and state agencies as well as other stakeholders. Some challenges mirror those for curtailing introductions: developing effective control strategies and finding the resources to implement them. Others are unique to spread within the U.S.: federal vs. state legal authorities, interactions among the agencies, free trade throughout the country, the highly developed transportation network, and the differing perceptions of risk among the several players. It is also unclear who can act if a state refuses to address a threat to its own resources. I summarize three approaches: APHIS leadership (re: emerald ash borer), APHIS/state collaboration (re: sudden oak death), and state independent actions (re: thousand cankers disease). I also reference several “orphan” pests and pathways.

The Black Oak Gall Wasp on Cape Cod and Martha's Vineyard

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ABSTRACT

Black oak, *Quercus velutina*, is the dominant deciduous tree on Cape Cod and Martha's Vineyard. In recent years, oaks on the cape and the islands have experienced severe canopy loss due to the infestation of the black oak gall wasp (*Callirhytis ceropteroides* (Bassett)). Oaks (*Quercus spp.*) in the region are under stress from previous winter moth (*Operophtera brumata*) defoliation and grow in low-nutrient sandy soils. These factors make them more vulnerable to gall wasp infestation. Little is known about the lifecycle of *C. ceropteroides* and the taxonomy of the species is still unclear. Our research aims to investigate the lifecycle of the black oak gall wasp, specifically emergence patterns, generation time, oviposition location, host specificity and survival rate. We will also perform DNA extraction, PCR analysis and sequence the CO1 barcoding gene for this species. Using the CO1 gene, we hope to determine if *C. ceropteroides* is the same species that caused extreme canopy loss on Long Island in the 1990s.

Systemic injections are commonly used to manage other gall wasp species, and may mitigate the black oak gall wasp infestation on Cape Cod and Martha's Vineyard. With other cooperators, we will evaluate the efficacy of trunk injections of emamectin benzoate and imidacloprid as control agents of the gall wasp on both low and heavily infested black oaks. The loss of a tree species that is known for its aesthetic value in residential communities and for wind protection in coastal areas will have both negative ecological and economic impacts on the region. These impacts present a need for integrated pest management. Our research could lay the foundation for future biological control efforts directed at this species. We hope our research will help arborists and landowners make future management decisions and will give the Cape Cod region one or more control solutions for the devastating and costly black oak gall wasp infestation.

Buying Time: Potential Role of Biocontrol in the Recovery of Native Vegetation After a Pest Invasion

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ABSTRACT

We are evaluating the impact of emerald ash borer (EAB) and its biocontrol on forest ecosystems. Although the effects of biocontrol on a forest pest are usually measured for the affected species only, these effects can propagate through the entire community. In the case of EAB, mature ash trees (*Fraxinus* spp.) are preferentially attacked by the pest, resulting in high mortality of this size class. As a result, the usually abundant ash seedling bank responds rapidly to openings in the canopy and populates the sapling layer. If these saplings are then affected by EAB, other fast-responding species, many of them weedy and invasive, can take over the ground and sapling layers and outcompete other slower growing, usually native, species.

However, if EAB biocontrol agents are being released into a particular stand, the sapling layer of ash trees may persist longer, buying time for the native community to respond to the new conditions before weedy and invasive species proliferate. To test these dynamics, we carried out a pilot study and collected vegetation data on forest stands affected by EAB in 2013. Study sites were chosen in areas affected by EAB without biocontrol and areas where two EAB biocontrol agents, *Tetrastichus planipennis* and *Oobius agrili*, are known to be established and spreading since their first release up to seven years ago. Stands were geo-referenced and information was collected on soil types, ecoregions, and percent of forested area around each site. Using release data, distance between plots, and vegetation surveys, we analyzed the effect of EAB biocontrol on the recovery of native vegetation. Biocontrol was found to have a significant positive effect on the presence of ash saplings. The abundance of ash saplings was associated with the percent of forested area around the stand, and it had a significant positive effect on the abundance of native seedlings. The percent cover of weedy/invasive groundcover had a significant negative effect on native species recruitment. Even if it does not prevent further EAB infestation, our pilot study indicates that the release of biocontrol agents can have a positive effect on the recovery of vegetation.

Investigating the Effects of Maternal Immune Priming on the Efficacy of a Fungal Biological Control Agent

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ABSTRACT

Asian longhorned beetles, *Anoplophora glabripennis* (Motschulsky), are wood borers native to China and Korea that were initially detected in North America in 1996. Breeding populations have been found in five eastern states in the US, one province in Canada, and six European countries. *A. glabripennis* has the potential to cause extensive damage in urban and natural forests if it becomes established in North America. The entomopathogenic fungus *Metarhizium brunneum* (Petch) is pathogenic to *A. glabripennis* and is being developed as a means for biological control.

Insects have an innate immune system characterized by no immune memory of a prior pathogen challenge and generating a generalized non-specific response to pathogen challenges. However, recent studies on the beetles *Tenebrio molitor* (L.) and *Tribolium castaneum* (Herbst) as well as bumble bees (*Bombus terrestris* L.) suggest that maternal pathogen exposure can subsequently increase offspring immunity and/or survival. We were interested in determining if a prior maternal pathogen exposure could make *M. brunneum* less effective in controlling *A. glabripennis* offspring. Specifically, we wanted to determine whether offspring of challenged mothers will live longer when exposed to a lethal pathogen challenge and how the method of maternal challenge (a live or a heat-killed challenge) will affect offspring immunity.

We investigated whether or not maternal exposure to the fungal pathogen *M. brunneum*, the closely related fungal pathogen *M. anisopliae* (Metsch.) or the gram negative bacteria *Serratia marcescens* (Bizio), would increase the immunity or survival of offspring challenged with a lethal dose of *M. brunneum*. We found that maternal exposure to some but not all pathogens increased offspring survival and we report the first instance of maternal priming providing protection against a fungal pathogen. This protection was not specific, as maternal exposure to the gram negative bacteria *S. marcescens* also enhanced offspring survival. Additionally, we found that the method of maternal exposure to a fungal pathogen mattered. Exposure to live *M. brunneum* increased offspring survival but exposure to dead *M. brunneum* did not increase offspring survival. Our findings suggest maternal exposure to the fungal biological agent *M. brunneum* will most likely have no impact on use of this pathogen as a biological agent because offspring survival was only slightly increased.

Cascade Effect of Laurel Wilt Disease on the Palamedes Swallowtail Butterfly in the Southeastern United States

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ABSTRACT

Laurel wilt disease (LWD) has caused widespread mortality of redbay (*Persea borbonia* (L.) Spreng.), a common understory tree of southeastern U.S. maritime forests. Redbay is the primary larval food source for the palamedes swallowtail butterfly (*Papilio palamedes* Drury) and LWD-induced mortality of redbay may have a direct effect on palamedes abundance. To determine if the removal of redbay has a direct effect on the abundance of the palamedes swallowtail, ~400 meter (1/4 mile) "Pollard transects" were walked and butterflies within 15 meters on each side of the observer were tallied. Three transects were walked per stand type (i.e. infected and uninfected stands). Infected stands are defined where LWD has been present ³3 years. To date, transects have been performed in Mississippi (2 years) and North Carolina (1 year) with data collection continuing for a minimum of 3 years in each state. Weather conditions (wind speed, temperature, % cloud cover, and humidity) were recorded at the beginning of each transect. Preliminary results from Mississippi indicate palamedes abundance has decreased sevenfold in infected stands, and in North Carolina palamedes abundance has decreased fourfold in infected stands. These results suggest that LWD-induced mortality of redbay is leading to a decrease in palamedes abundance and extinction or long term decline of redbay may lead to the loss of the palamedes swallowtail throughout much of its range. Other multi-trophic cascade effects may also become evident as LWD continues to alter the ecosystem. These results indicate the need for focused conservation efforts aimed at protecting crucial palamedes habitat and planting and management of refugia. These results also indicate the need to monitor the removal of sassafras, another laurel species vulnerable to LWD, and its possible effect on spicebush swallowtail (*Papilio troilus* L.) abundance.

Canaries in the Coalmine: Urban Warming Predicts Climate Change Effects on Forest Pests

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ABSTRACT

Predicting the effects of climate change on forest health is critical to protect forest resources, biodiversity, and ecosystem services. Current options to study climate change effects include experimental warming experiments and space for time substitutions such as latitudinal gradient experiments. Unfortunately, the small scale and great expense of warming experiments and confounding variables associated with latitudinal and altitudinal gradients limit their predictive power. For centuries cities have had many of the atmospheric changes predicted to occur worldwide under climate change. In particular, cities can be 10° C warmer than surrounding areas. We predict that urban warming will increase tree pest abundance and fitness and that these changes are congruent with effects of warming in natural forest ecosystems. If so, cities could serve as replicated study sites from which to predict the consequences of global warming.

Many herbivorous arthropods, particularly tree pests, are more abundant and damaging in urban than rural areas. Urbanization has many effects on ecosystems but the urban heat island effect is one of the most pervasive. However, no research has examined how urban heat affects the fitness and abundance of herbivorous arthropods. We predict that warm habitats increase herbivore abundance due to direct physiological benefits and a breakdown of trophic dynamics. We test this hypothesis with survey and manipulative experiments that focus on two common scale insect pests, *Parthenolecanium quercifex* (Hemiptera: Coccidae) and *Melanaspis tenebricosa* (Hemiptera: Diaspididae) and their natural enemies. We sampled scale insects on red maple (*Acer rubrum*) and willow oak (*Quercus phellos*) trees in the hottest and coolest parts Raleigh, NC USA. We overlaid a surface temperature map of each city with a map of street trees in Arc GIS. We visited 20 willow oak and 30 red maple trees in the hottest and coolest pixels of the map and counted *P. quercifex* and *M. tenebricosa* on 4 branches per tree. We dissected scale insects to record size, fecundity, and phenological stage on each sample date.

The hottest trees in Raleigh had up to 300 times more scale insects than trees just 2 degrees cooler. At hot sites, scales were also larger, more fecund, and had greater survival. Moreover, an analysis of all red maple street trees in Raleigh found that those in the hottest sites were twice as likely to be in poor condition and half as likely to be in excellent condition than trees in cooler sites.

To test the hypothesis that cities predict the effects of warming in rural forests we examined the response of *M. tenebricosa* to temperature in the context of an urban heat island, a series of historical temperature fluctuations, and recent climate change. We surveyed *M. tenebricosa* on 55 urban street trees in Raleigh, NC; 342 herbarium specimens collected in the rural southeastern United States from 1895 to 2011, and at 20 rural forest sites represented by both modern (2013) and historical samples. We related

scale insect abundance to August temperatures and found that *M. tenebricosa* was more common in the hottest parts of the city, on historical specimens collected during warm time periods, and in present-day rural forests compared to the same sites when they were cooler. Peak scale insect abundance occurred at comparable temperatures in the urban and historical datasets. However, maximum insect abundance was ~6 times greater in the city than in rural areas. The qualitative congruence in our results suggests that *M. tenebricosa* may become increasingly common in some rural forests as a result of global warming.

Effects of Urban Forest Fragmentation on Abundance, Diversity, and Health of Native Bees

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ABSTRACT

Currently, farmers rely heavily on managed pollinators such as honey bees, bumble bees, and mason bees, which represent less than 1% of the total 20,000 – 30,000 species of bees. With overexploitation and an observed decrease in the health of managed pollinators, current research is demonstrating that native bee communities can be used as an insurance policy for managed bees. Although honey bees pollinate a vast number of plants, native pollinators display unique foraging behaviors and are equally as important. Therefore, it is imperative to understand their life histories and the associated ecosystem services they provide. Specimens were sampled from 15 sites in Northern Delaware and Southeastern Pennsylvania associated with the university-wide FRAME (Forest Fragments in Managed Ecosystems) project using both bowl traps and aerial sweep netting. Anticipation of declining honey bee populations requires research to find alternatives for the pollination industry. Native pollinators are plentiful in many ecosystems and provide as much pollination as managed bees in some agricultural settings, and additionally continue to provide vital pollination services to native fauna.

Microsclerotia Applied in Hydromulch to Control Asian Longhorned Beetles

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ABSTRACT

Recently it was discovered that the entomopathogenic fungus *Metarhizium brunneum* F52 (Hypocreales: Clavicipitaceae) is able to produce environmentally persistent microsclerotia (compact fungal hyphal aggregations). Incorporating these desiccation-tolerant microsclerotia (Mb MS) granules into hydromulch [a mixture of water + wheat straw mulch (Hydrostraw™) + psyllium tackifier], represents a novel, easy-to-use and environmentally-friendly mycoinsecticide that can be sprayed onto the trunks of forest or orchard trees to potentially control pests. Hydromulch absorbs environmental moisture that allows microsclerotia to germinate, and the production of spores in turn, causes lethal infections in Asian longhorned beetles, *Anoplophora glabripennis* Motschulsky (Coleoptera: Cerambycidae). To test how quickly beetles could be killed, moist and dry bark pieces and filter paper were sprayed with a low dose (~9 Mb MS/cm²) of microsclerotia in hydromulch. Median survival times of beetles exposed to moist bark and filter paper were 21.5 days and 23.5 days, respectively. Beetles exposed to dry bark died significantly slower ($\chi^2_3 = 33.9$; $P \leq 0.0001$) with a median survival time of 35.5 days. This highlighted the importance of moisture on the substrate so that microsclerotia will germinate. In an attempt to kill beetles faster, moist bark pieces were sprayed with three doses of microsclerotia in hydromulch: low (6-9 Mb MS/cm²); medium (10-19 Mb MS/cm²) and high (20-30 Mb MS/cm²). At high doses, 50% of beetles died in 12.5 days but at lower doses it took significantly ($\chi^2_3 = 24.39$; $P \leq 0.0001$) longer to kill beetles (16.5 days-18.5 days). Outdoor persistence studies, using moist bark pieces, sprayed with a high dose of hydromulch (20-30 Mb MS/cm²) and attached to four trees in the woods were undertaken. Every 4 days, bark pieces were removed from trees and the numbers of spores produced were counted for up to 24 days. Sub-sampling was repeated for another 24 days. There was a significant increase ($\chi^2_{11} = 276.22$; $P \leq 0.0001$) in spore production over the 24 days and in the second replicate (another 24 days) spore production was significantly higher than in the first replicate ($P \leq 0.0001$). Importantly, rainfall was significantly correlated ($P \leq 0.0042$) to this increase in spore production in the second replicate. Environmental moisture and to a lesser degree, temperature plays a big role in the spore production by microsclerotia and will subsequently affect the levels of insect mortality.

Alliance for Saving Threatened Forests: Breeding for Resistance to Hemlock Woolly Adelgids

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ABSTRACT

Hemlock woolly adelgids (HWA; *Adelges tsugae*) were introduced into the eastern United States in the mid-twentieth century. HWA is devastating eastern hemlock (*Tsuga canadensis*) and Carolina hemlock (*T. caroliniana*) ecosystems and the ornamental hemlock trade. The long-term goal of the Alliance for Saving Threatened Forests is development of trees true-breeding for resistance to adelgids. Resistant hemlocks would be suitable for use in the ornamental industries and for restoration of affected natural eastern hemlock and Carolina hemlock stands throughout their affected ranges. We are exploring a variety of approaches to incorporate resistance into deployment populations. Breeding resistant genotypes from native populations is a priority, as is crossing native species with potentially resistant exotic species to produce interspecific hybrids, and then backcrossing resistant hybrids with native species. We will study mechanisms of resistance and inheritance to develop novel methods for resistance screening, selection, and gene incorporation. Our selection and breeding strategy includes: 1) Identify genotypes in natural stands putatively resistant to or tolerant of adelgids. 2) Through rooted cuttings and grafting, establish putatively resistant genotypes in several common garden plantations. 3) Evaluate resistance or tolerance through artificial infestation with adelgids. 4) Breed selected genotypes and test offspring for resistance or tolerance. For interspecific hybrids we will take advantage of the potential for genetic resistance or tolerance in other species of hemlock. Exotic hemlocks we are attempting to cross with eastern and Carolina hemlock include Chinese (*T. chinensis*), N. Japanese (*T. diversifolia*), S. Japanese (*T. sieboldii*), and western (*T. heterophylla*). Where crosses produce viable seed, progeny will be tested for resistance or tolerance in greenhouses and common gardens using artificial infestations. Resistant progeny will be backcrossed with the native species. Somatic embryogenesis may be utilized to produce larger numbers of hybrid ramets, and to attempt embryo rescue where crossing barriers may exist between eastern and Asian hemlocks. To date, ~50 native hemlock trees are in the resistance/tolerance testing pipeline. Carolina and eastern hemlock were each crossed with Chinese, northern Japanese, southern Japanese, western, Carolina, and eastern hemlock in 2011-2013. Pollen for the Asian hemlocks crosses was obtained from sources at Longwood Gardens, the U.S. National Arboretum, and University of Rhode Island. Carolina by Chinese hemlock hybrids produced by the U.S. National Arboretum are being field grown in NC to evaluate adaptability and ornamental traits. Dr. Scott Merkle at University of Georgia has initiated cultures for somatic embryogenesis with hemlock hybrid crosses between the Asian hemlocks and both eastern and Carolina hemlock.

Local Conditions Influence Gypsy Moth Outbreak Regulation by *Entomophaga maimaiga*

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ABSTRACT

Population outbreaks of the non-native forest defoliating gypsy moth, *Lymantria dispar*, often occur in periodic cycles, particularly in the US mid-Atlantic region. Outbreking populations generally collapse after 1-3 years due to regulation by the fungus *Entomophaga maimaiga* and a nucleopolyhedrovirus (*LdNPV*). Because *E. maimaiga* and *LdNPV* are both sensitive to environmental conditions, we examined the potential roles of a suite of factors in driving *E. maimaiga* epizootics at 63 mid-Atlantic sites during the final year of a synchronous gypsy moth outbreak. *Entomophaga maimaiga* was the most prevalent natural enemy while *LdNPV* prevalence was much lower. *E. maimaiga* and was locally spatially aggregated and prevalence of fungal infection was associated with host density. *LdNPV* prevalence was also associated with host density but the virus was not spatially aggregated. Levels of infection for both pathogens were associated with environmental moisture levels although associations were not easy to interpret. *Entomophaga maimaiga* levels could not be predicted based on physiographic conditions although *LdNPV* prevalence was associated with some physiographic characteristics of sites. During this study, parasitoids were also recorded but levels of the four species collected (the braconid *Cotesia melanoscela*, the ichneumonid *Phobocampe uncinata* and the tachinids *Parasetigena silvestris* and *Compsilura concinnata*) were generally low, and not associated with host density.

Native and Non-native Cerambycid Beetles in Urban Forest Fragments

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ABSTRACT

We are evaluating the abundance and species richness of beetles in the family Cerambycidae in forest fragments in northern Delaware. These urban and suburban forests are part of the long-term Forest Fragments in Managed Ecosystems (FRAME) program, which comprises 30 sites in the Mid-Atlantic region of the eastern United States. All sites, including those used for this study, have been characterized in regards to soil, understory vegetation species and density, litter dwelling arthropods, and occurrence of amphibians, reptiles, birds, and mammals. The cerambycid family is of particular interest because it contains a number of native and nonnative forest pests. Many of these can be brought to baited traps. We used a combination of three synthesized aggregation pheromones and ethanol as our bait at 24 different sites. Of the 24 sites, six of them had two traps, for a total of 30 traps. We collected all of the insects captured in these traps once a week from April 30 to September 26, 2013. The data gathered during this field season show that each species of cerambycid is impacted differently by differences in the forest fragment composition. The most important factors for influencing cerambycid occupancy and abundance are tree species and time of year. We are currently running more analyses to discover more drivers behind abundance patterns.

Creation and Validation of a Degree-Day Model for *Sirex nigricornis* f. (Hymenoptera: Siricidae) in Arkansas

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ABSTRACT

Sirex nigricornis F. (Hymenoptera: Siricidae) is a pine-inhabiting woodwasp native to the eastern United States and Canada. It is not known to attack healthy pine trees and is not normally considered a pest. However, *S. noctilio*, a non-native woodwasp introduced into the northeastern U.S. in 2004, has been shown to attack and kill healthy trees. The Southeastern U.S. contains millions of hectares of economically, and ecologically valuable pine stands. These stands are at risk if, or when, *S. noctilio* reaches the Southeast. We created a degree-day model for *S. nigricornis* and compared those data to previously published information on *S. noctilio*. Several authors report *S. noctilio* emergence beginning at 2,500 cumulative degree-days (CDD) or earlier, regardless of the region in which the woodwasps are being studied. This is in contrast to the 4,000 or more CDD required for *S. nigricornis* emergence in Arkansas. This vast difference in CDD requirements suggests that *S. noctilio* emerge earlier and have access to host material far before emergence of native woodwasps in the Southeast. They could potentially create additional host material for other pine-inhabitants by attacking healthy trees earlier in the year or, conversely, they could displace native species by utilizing available resources. Future experiments will involve examining mortality caused by *S. nigricornis* parasitoids and nematodes.

Sex-Specific Trail Pheromone of the Asian Longhorned Beetle, *Anoplophora glabripennis*

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ABSTRACT

Anoplophora glabripennis is a polyphagous member of the Cerambycidae, which is considered world-wide to be one of the most serious quarantine pests of deciduous trees. We isolated and identified four chemicals from the trail wash of *A. glabripennis* virgin and mated females that were not present in those of males. These compounds were identified as 2-methyldocosane and (Z)-9-tricosene (major components), as well as (Z)-9-pentacosene and (Z)-7-pentacosene (minor components); every trail wash sample contained all four chemical components, although the ratios changed with age of the female. Males responded to the full pheromone blend, regardless of their mating status, but virgin females avoided the pheromone, suggesting that these females may use it as a spacing pheromone to avoid intraspecific competition and maximize resources. Virgin, but not mated, males also chose the major pheromone components in the absence of the minor components over the control. Taken together, these results indicate that all four chemicals we identified are components of the trail pheromone. The timing of production of the ratios of the pheromone blend components that produced positive responses from males coincided with the timing of sexual maturation of the female.

Biological Control of Emerald Ash Borer in Maryland: An Update on Parasitoid Recovery and Establishment

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ABSTRACT

Emerald ash borer (EAB), *Agrilus planipennis* Fairmaire, was first detected in Maryland in 2003 and has since spread to 11 counties in the western half of the state. After initial eradication efforts were unsuccessful, a biological control program was initiated in 2009 involving the release of two larval parasitoids (*Spathius agrili* and *Tetrastichus planipennisi*) and one egg parasitoid (*Oobius agrili*) from the native range of EAB. As of 2013, a total of 212,273 parasitoids have been released in Maryland, comprising 55,557 *S. agrili*, 144,219 *T. planipennisi*, and 12,497 *O. agrili*. These parasitoids have been released at 34 locations across the state. Beginning in 2010 and continuing annually, parasitoid recovery has been quantified in the field by debarking EAB-infested ash trees (*Fraxinus* spp.) to observe egg and larval parasitism each spring and/or fall. Additionally, from 2012 onwards we have monitored parasitoids recovered from field-collected ash logs stored in rearing barrels. Debarking of ash trees has recovered a total of 90 EAB larvae parasitized by the two introduced larval parasitoid species, with nine introduced parasitoids recovered from rearing barrels (which we are unable to confirm emerged from EAB larvae). Further, field surveys of EAB eggs showed that the egg parasitoid (*O. agrili*) was recovered in two locations. We have also recovered 48 EAB larvae parasitized by the native parasitoid species *Atanycolus* spp. and *Spathius floridanus*, and the accidentally introduced species *Balcha indica*, with an additional 65 individuals of these three species collected from rearing barrels. In summary, all three introduced parasitoids have been recovered in Maryland, but the vast majority (81.1%) has been *T. planipennisi*. These parasitoids have been recovered from 17 locations, and *T. planipennisi* has been recovered from four locations where no releases occurred which suggests that this species is successfully establishing and dispersing. Future plans include the potential release of *Spathius galinae* (pending approvals) to further augment the EAB biological control program in Maryland.

Effects of Temperature on Neonate *Lymantria* Survival With and Without Food

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ABSTRACT

Females of *Lymantria dispar*, Asian gypsy moth, and *L. monacha* (nun moth) can fly and are attracted to lights at night. In well-lit Asian ports near forests females lay their egg masses on the superstructure and cargo of ships at night. If an egg mass on a ship hatches while the ship is in another port, the neonates could disperse, find suitable hosts, and establish. What is unknown is how long the neonates can survive at different temperatures without food or if the only food they find is either at the wrong phenological state or from a less preferred host. To address this data gap, larval survival and development were assessed at seven temperatures: 1, 5, 10, 15, 20, 25, and 30°C. At each temperature, 50 newly hatched larvae from each of 9 strains (3 European and 5 Asian *L. dispar*, and 1 *L. monacha* strain) were established on each of the following treatments: no food, Douglas-fir foliage (*Pseudotsuga menziesii* var. *menziesii*), and black oak foliage (*Quercus velutina*). Larvae with no food were held singly and checked daily until death. Those on foliage were held in groups of 10, the foliage was changed as needed, larvae were checked daily for 14 days for survival and molts, and survivors were weighed at 14 days.

The number of days a neonate could survive without food decreased with increasing temperature between 5 and 30°C. At 5°C, more than half of the larvae survived more than a month; at 30°C, most were dead within the first week; and at 1°C, all larvae died within two weeks. There was some obvious *L. dispar* feeding on both foliage types at 10°C but the *L. monacha* larvae fed more, especially on the Douglas-fir. At the higher temperatures, larvae that fed generally grew and molted successfully on both foliage types, some reaching the second instar in 14 days at 15°C, and many reaching the fourth instar at 30°C. There was considerable variation in the survival and development of larvae from both European and Asian *L. dispar* strains on Douglas-fir. These results suggest that if larvae were to hatch from egg masses that are moved through the transportation system during times of the year when the temperatures are between 1 and 20°C they will have a week or more to find food and could establish if the foliage they find is adequate. At temperatures above 20°C, larvae will have a shorter period of time to find adequate food but there may be a greater variety of foliage types available during the time of the year when those temperatures would occur.

Field Observations of *Sirex Noctilio* Mating and Male Aggregation Behavior

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ABSTRACT

The European woodwasp, *Sirex noctilio* Fabricius (Hymenoptera: Siricidae), is a significant pest in forests where it has been accidentally introduced. In 2004, the woodwasp was found to be established in northern New York state and southern Ontario, and has since spread steadily through the northeastern United States and adjacent regions in Canada. *S. noctilio* males are known to aggregate in swarms above pine forest canopies, where they mate with visiting females. Laboratory experiments have demonstrated the presence of a contact pheromone in female wasps and a volatile pheromone in males. Because males are rarely observed near the ground in pine stands, few details of their lekking have been observed, and the effects of these pheromones on wasp behavior have never been tested in the field.

In November 2013, *S. noctilio* field observations in a *Pinus patula* plantation in northern Mpumalanga province, South Africa, confirmed previous reports of the presence of male leks and the probable role of a contact pheromone during mating. Males aggregated at the high points of log piles in groups of nine to 48 individuals, and responded rapidly to artificial manipulation of apices. Male activity levels rose during sunny conditions and aggressive behavior increased in the presence of female wasps. These observations suggest that forest structure and variable weather, along with clear positive phototaxis, influence lek formation. This may help explain poor trapping success rates and suggest strategies for the development of better techniques.

***Scymnus camptodromus* Development and Predation of Hemlock Woolly Adelgid**

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ABSTRACT

The hemlock woolly adelgid is an invasive insect pest that threatens the sustainability of eastern hemlock forests. This insect is capable of causing severe tree decline, has no known parasitoids and is the single most important threat to native hemlock stands in the eastern United States. To reduce damage on hemlocks *Scymnus camptodromus*, a specialist predator of HWA, was brought to the US from China as a potential biological control agent. This beetle has been approved for removal from quarantine but is not yet approved for field release. Larvae of *S. camptodromus* have been reported to be voracious predators of HWA, but their development and prey requirements have not been studied. We evaluated *S. camptodromus* larval development time and predation by instar, strain and temperature. We observed that temperature had significant effects on the predator life history. (1) the larvae tended to develop faster and consume more HWA eggs per day as the temperature increased. Mean HWA egg predation per day was less at 15 °C (22.9 ± 0.8) than at 20 °C (31.4 ± 0.9). Although, larvae ate less per day at 15°C temperature, they also took longer to develop to adults at this temperature, resulting in a greater number of eggs consumed during larval development at 15 compared to 20 °C; and (2) the temperature thresholds of predator larval development lies between 0-25 °C, which closely matches the development thresholds of HWA progrediens. Confined release field studies of *S. camptodromus* adults over the field seasons of 2012 and 2013 showed a significant reduction in HWA populations. In 2012 HWA egg reduction was 12% more with *S. camptodromus* adults compared with the natural reduction in unbagged branches. Similarly, HWA egg reduction was 27% more with the predator over an 8-week period during the field season in 2013. Also in 2013, there was a 35% reduction in the number of HWA settled crawlers in bags with the predator compared to controls. Under the conditions of our studies, *S. camptodromus* showed potential to be an effective predator of HWA with close synchrony to the HWA life cycle.

Attraction of Non-Cerambycids to Cerambycid Pheromones in Southeastern USA

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ABSTRACT

Numerous hardwood borers (Coleoptera: Cerambycidae) are broadly attracted to blends of hydroxyketones and hexanediols (Hanks et al. 2012, Can. J. For. Res. 42: 1050-1059). Our objective in this study was to determine if these compounds had any effect on the responses of other species, particularly ambrosia beetles. We conducted the same experiment once near Eatonton, GA during the spring of 2012 (22 March – 17 May) and again near Staffordville, GA during the summer of 2013 (10 July – 20 August). Ethanol lures and lures of racemic hexanediol (C6-diol), racemic 3-hydroxy-2-hexanone (C6-ketol) and racemic 3-hydroxy-2-octanone (C8-ketol) were supplied by Con-Tech Inc. (Delta, BC). The hydroxyketones were provided by Bedoukian Research Inc. (Danbury CT) whereas hexanediols were synthesized in Fredericton NB. The release rates of the diols and ketols ranged from 10-40 mg/d depending on temperature whereas ethanol released at about 0.6 g/d.

In both experiments, we used 10-unit multiple-funnel traps that were modified by increasing the center hole of each funnel from 5 cm to 12 cm, thereby allowing placement of all lures within the confines of the trap. At each of the two locations, we deployed 40 funnel traps, set in 10 replicate blocks of 4 traps each. Traps and blocks were spaced 8-12m apart. One of the following treatments was allocated to a trap within each block: (1) ethanol alone; (2) ethanol + C6-diol; (3) ethanol + C6-ketol; and (4) ethanol + C8-ketol. Lures were replaced once after three weeks. All cups contained a solution of propylene glycol.

The bark beetle predators, *Temnoscheila virescens* (Trogossitidae) was attracted to 3-hydroxy-2-hexanone whereas *Chariessa pilosa* (Cleridae) was attracted to the hexanediols. The bee assassin bug, *Apiomerus crassipes* (Hemiptera) was attracted to both hydroxyketones. Attraction of the ambrosia beetles, *Dryoxylon onoharaensis* (Curculionidae: Scolytinae) to ethanol was interrupted by 3-hydroxy-2-octanone. Catches of four other species were unaffected by the hexanediols and hydroxyketones. The red-shouldered bostrichid, *Xylobiops basilaris* was attracted to the hexanediols. Catches of *Buprestis* spp. (Buprestidae) and *Hylobius pales* (Curculionidae) were unaffected by the cerambycid pheromones. The bark beetle, *Hypothenemus rotundicollis* (Curculionidae: Scolytinae) was attracted to 3-hydroxy-2-octanone.

Variation in Effects of Conophthorin on Attraction of Ambrosia Beetles to Ethanol-Baited Traps

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ABSTRACT

To date, more than 60 species of ambrosia beetles have invaded the United States with several species such as *Xylosandrus crassiusculus* and *X. germanus* causing economic damage in fruit orchards and horticultural nurseries. In several studies, conophthorin has been implicated as an attractant for ambrosia beetles. Our objective was to determine if conophthorin can significantly (and consistently) increase catches of various species of ambrosia beetles in the United States. In 2013, we conducted the same experiment at four locations across the USA: (1) Georgia (13 March – 9 May); (2) New Hampshire (3 May – 26 June); (3) Michigan (8 May – 1 July); and (4) Oregon (16 May – 11 July). Multiple-funnel traps and ethanol were supplied by ConTech Inc. (Victoria, BC). Conophthorin lures were obtained from ConTech for Georgia whereas Synergy (Burnaby BC) supplied the remaining conophthorin lures. The release rate of ethanol was about 0.6 g/d whereas conophthorin was released at about 1-2 mg/d. At each location, we deployed 20 funnel traps, set in 10 replicate blocks of 2 traps per block. Traps were spaced >10m apart. One of the following treatments was allocated to a trap within each block: (1) ethanol alone; and (2) ethanol + conophthorin. All cups contained solution of propylene glycol.

Conophthorin decreased catches of *X. germanus* in Oregon but had no effect in Michigan and New Hampshire. Conophthorin increased catches of *X. saxesenii* in New Hampshire and Oregon but had no effect in Georgia and Michigan. Conophthorin increased catches of *Cyclorhipidion pelliculosum* in New Hampshire but had no effect in Michigan. Conophthorin had no effect on catches of *Ambrosiophilus atratus*, *Anisandrus dispar*, *A. sayi*, *Dryoxylon onoharaensum*, *Gnathotrichus sulcatus*, *Monarthrum fasciatum*, *M. mali* and *Xylosandrus compactus*. In Georgia, conophthorin increased catches of *G. materiarius*, *Xyleborus* spp. and *X. crassiusculus* but decreased catches of *Cnestus mutilatus* and *Cyclorhipidion bodoanum*.

Drought as a Predisposing Factor for Bark Beetle Attack

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ABSTRACT

Drought periods induce a continuously increasing water stress in trees which - under Central European conditions –become more and more susceptible as hosts for aggressive bark beetle species, like the spruce bark beetle *Ips typographus* (Christiansen and Bakke, 1997). As it is known from various conifers, this indirect effect is due to a lower tree defense reaction resulting from reduced photosynthesis, lower oleoresin monoterpene content (Levinsohn et al., 1991) and a decrease in resin flow (Reeve et al., 1995). The aim of this interdisciplinary project is to find thresholds for the interaction between soil and tree water balance and the susceptibility of the host tree for bark beetle attack which can be used to quantify the actual risks of drought as a predisposing factor for bark beetle attack.

At six experimental plots (size: 18x20 m each) in an upper, moderately north-west exposed, 90-year old Norway spruce stand mixed with European beech, a roof construction was used to establish a gradient of water deficit. Therefore, two of these plots were totally covered, two plots were partially covered, and two plots were uncovered and serve as controls. From each plot, three spruce trees were selected for tree physiological analyses and induced attack experiments with bark beetles.

In multivariate data analysis we test the hypothesis that a relationship exists between increased soil water deficit, reduced tree defenses and an increased susceptibility of the tree for bark beetle infestation. Combined with the hydrological “Coup Model”, which is based on the climate data, forest stand and soil-related parameters of the study site (Jansson and Karlberg, 2010), the results will allow us to estimate the predisposing effect of drought for bark beetle attack.

Furthermore, the field data will be combined with modelled data of the phenology model “PHENIPS” (model for estimating the development of *Ips typographus*, Baier et al., 2007) and the Predisposition Assessment System “PAS” (Netherer and Nopp-Mayr, 2005) in order to obtain a dynamic model for risk assessment of site and stand-related conditions for bark beetle outbreaks.

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Conservation of Ash Diversity: Effects of Wide-scale Insecticide Application on Genetic Structure, Recruitment, and Regeneration of Ash in Forests Invaded by Emerald Ash Borer

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ABSTRACT

Wide-scale mortality of *Fraxinus* (ash) by emerald ash borer (*Agrilus planipennis*, EAB) threatens the genus with regional extirpation, creating the need for strategies to conserve the ash gene pool. In such an attempt, the Five River MetroParks (FRMP) system near Dayton, OH initiated an ongoing program to protect 600 mature ash trees with insecticide. Their overall goal was to pre-emptively protect a proportion of ash within the FRMP park system, so they selected a proportion of each of the five ash species (*F. americana*, *F. pennsylvanica*, *F. quadrangulata*, and *F. profunda*) approximately equal to the proportion of species abundance to treat with insecticide. Our goal is to evaluate the effectiveness of this conservation program on genetic diversity and ash population dynamics. We hypothesize that through the protection of mature, reproductive ash, stable population structures (i.e., structure supporting regeneration and recruitment) and genetic variation can be maintained during the EAB invasion. We are characterizing the demography of different ash age cohorts in order to determine the effects of 1) EAB invasion and 2) insecticide treatments on ash density, survival, reproduction, and genetic variation. Ultimately, these data will be used to generate an optimized model of ash conservation via selective insecticide treatment that can be used to conserve phenotypic and genetic variation in other ecosystems invaded by EAB.

Ash Mortality Caused by Emerald Ash Borer Impacts Forest Floor Invertebrate Communities

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ABSTRACT

Emerald ash borer (EAB; *Agilus planipennis*) is an invasive wood-boring beetle native to Asia. Since its introduction into North America, EAB has killed untold millions of ash trees, resulting in widespread simultaneous formation of canopy gaps in forests. Gap formation may alter environmental conditions on the forest floor, potentially impacting populations of ground-dwelling invertebrates. However, as dead ash trees fall, accumulation of coarse woody debris (CWD) may buffer environmental changes on the forest floor induced by increased light. The objective was to quantify the effects of gap formation and accumulation of CWD caused by EAB-induced ash mortality on forest floor invertebrate community diversity and composition. A two-year, manipulative experiment was conducted in forests at NASA Plum Brook Research Station in Ohio to determine the effects of light gaps, CWD, and their interaction on forest floor invertebrates. Canopy gaps altered the invertebrate community by decreasing family and guild richness in 2011. However, the effect of gaps varied between years, and was marginal in 2012. Invertebrate community composition shifted as important members of predator and detritivore guilds were more prevalent under closed canopy. Although gaps increased soil temperature and soil moisture overall, the difference in environmental conditions between gaps and closed canopy habitat was greater in 2011 than 2012. Invertebrate community response was dependent on the degree of environmental differences between habitats. CWD had no effect on forest floor invertebrates. EAB indirectly impacted forest floor invertebrates through changes in environmental conditions on the forest floor caused by ash mortality.

The U.S. Asian Gypsy Moth Pest Exclusion Program: Monitoring and Ship Inspection Programs in Japan, Korea, China, and the Russian Far East

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ABSTRACT

The Asian gypsy moth has been traditionally used to refer to any of the biotypes of *Lymantria dispar* possessing female flight capability. It includes the following species and subspecies: *L. dispar asiatica*, *L. dispar japonica*, *L. albescens*, *L. umbrosa*, and *L. postalba* (Pogue and Schafer, 2007). For this program, the risks of the introduction of other *Lymantria* species whose females can fly and are attracted to light, such as *L. xyliana*, must also be taken into consideration. These insect species and subspecies have invasive biology and potential for significant economic damage, and have been found in port areas in the U.S. and Canada. Annual economic value of selected commodities that could be impacted by AGM is estimated to be around 18.4 billion US dollars. The following risk mitigation measures have been taken to prevent the introductions of AGM into the United States.

1. Population monitoring and management at foreign ports. Many foreign seaports within the AGM range are monitored. As early as 1993, a cooperative program was initiated between Russia and the U.S. to develop an early warning system and to reduce the risk of *Lymantriid* introductions from the Russian Far East into the U.S. and other participating countries. Japan and South Korea also have begun to participate in this program after AGM outbreaks and the subsequent contamination of ships arriving from these countries. The trapping results together with other related data provide information for determining risk period, and relevant population levels at different marine ports in different countries.

2. Inspection and certification of vessel at foreign ports. In 2009, the secretariat of the North American Plant Protection Organization (NAPPO) issued "Guidelines for Regulating the Movement of Ships and Cargo from Areas Infested with the Asian Gypsy Moth" (NAPPO RSPM #33). Inspection and certification of vessels for AGM at foreign ports have increased significantly since 2010, with Japan and Korea participating for the first time in 2010. Inspections in China began in 2011. The inspections have continued to increase in Japan, Korea, and China in 2012 and 2013, and the number of certificates issued has increased rapidly since 2010 (fig. 1). Special procedures for suspect AGM ships (APHIS PPQ 03/2012-139) for clearing vessels provides guidance and protocols to prevent the artificial spread of AGM from high-risk areas including Far East Russian, South Korean, Japanese, and selected Chinese seaports where AGM populations are at high densities.

3. Vessel inspection at the ports-of-entry in the U.S. and Canada. Port-of-entry inspections of vessels by Customs and Border Protection (CBP) and Canadian Food Inspection Agency (CFIA) provide quality assurance that offshore inspections are being carried out in a satisfactory manner. Another important piece of the exclusion program is the excellent compliance from industry. Many shipping

companies are putting protocols in place to comply with AGM entry requirements to avoid delays.

4. Survey at U.S. and Canadian ports and adjacent areas. Previous local introductions were found through surveys in both the U.S. and Canada in different years and in different states/provinces. In addition, there have been intensive European gypsy moth trapping programs across the U.S., and sample moths from the survey in the U.S. have been genetically analyzed, mostly by the USDA-APHIS-PPQ S&T Otis laboratory. These analyses should be able to detect any AGM, should it be present.

5. Collective Efforts and Outreach Activities. USDA-APHIS and CFIA have engaged counterparts in China, Japan, South Korea and Russia for mitigation measures, and as well as marine shipping companies, and provided training to personnel at AGM-free certification organizations.

We are reviewing AGM port monitoring data from different countries in 2013 and work with all concerned parties for better resource allocation to mitigate AGM risks. We will also conduct cooperative research on biology of AGM, especially the female flight behaviors, to better understand the risk factors, and improve techniques for surveying and monitoring as well as vessel inspection.

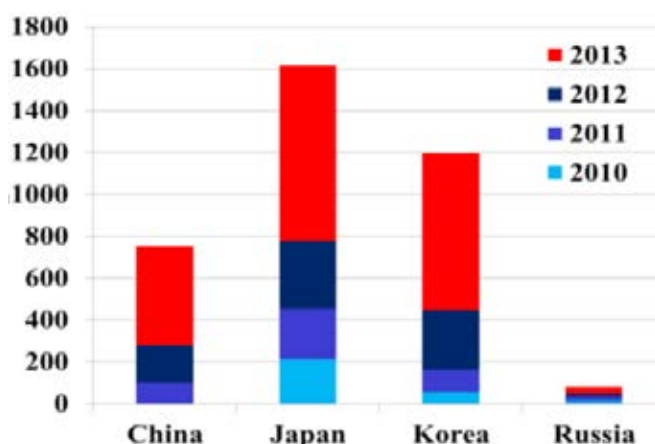


Figure 1 (left). AGM-free certification issued by different countries for vessels destined for the U.S.

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Recent Outbreaks of the Apple Jewel Beetle (Coleoptera: Buprestidae) in Northwest China: A Potential Risk to Apple Production

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ABSTRACT

The apple jewel beetle, *Agrilus mali* Matsumura (Coleoptera: Buprestidae), is an occasional minor pest of apple trees in north and central China and rarely causes any economic loss there. However, it has recently invaded the Xinjiang Uyghur Autonomous Region of northwest China, where it was first detected on cultivated apple trees (*Malus pumila*) in 1995 and had an outbreak on wild apple (*Malus sieversii*) trees in 1999. Entomologists in China suspected that the initial population of *A. mali* in Xinjiang had probably originated from immature stages infesting the young apple trees imported from Shandong Province (east coast of China) in 1993. *Agrilus mali* has now become a supplementary quarantine pest in several infested provinces of China, and is included into a National Forest Risk Pest List by the State Forestry Administration of China in 2013.

Agrilus mali has a distribution including Russia, Japan, Korea, and China. Based on published records, this beetle occurs in 15 provinces or municipal regions in China, including Heilongjiang, Jilin, Liaoning, Hebei, Henan, Beijing, Inner Mongolia, Shandong, Shanxi, Shaanxi, Gansu, Ningxia, Hubei, Qinghai, and Xinjiang. Approximately a dozen host plant species have been recorded in the literature as its food plants, which include *Malus pumila*, *M. sieversii*, *M. asiatica*, *M. prunifolia*, *M. yunnanensis*, *Prunus salicina*, *P. persica*, *P. armeniaca*, *Salix babylonica*, *Juglans regia*, *Ceraras* spp., *Pyurs* spp., and *Crataegus* spp.

Agrilus mali is univoltine in northwest China. It overwinters as a young larva under the bark of branches or trunks usually in early November, and the development of the insect is usually delayed 20-30 days in high altitude areas than normal conditions. Adults feed on edge of apple leaves, before mating and laying eggs 15-20 days after emergence. D-shaped exit holes remain on the branch or trunk as characteristic signs of emergence. Females lay 60-70 eggs during their lifetime, and eggs hatch in 10-13 days. The newly hatched larvae chew into the bark and reach the xylem as they develop. The phloem and cambium are severely damaged, and larval galleries are filled with brown frass. Some red or yellow sap may appear on the bark surface above an area infested with *A. mali* larva inside the branch or trunk. Canopy dieback normally occurs 2-3 years after infestation.

A total of 11 species of natural enemies of *A. mali* have recently been discovered, including 1 predator of the egg-larval stage, 7 larval parasitic wasps, 1 pupal ichneumonid wasp, 1 larval-pupal chalcid, and 1 parasitic mite. These natural enemies together have inflicted >33% mortality of *A. mali* in Xinjiang, China. Other biotic factors such as pathogenic microorganisms and tree resistance can also cause up to 30% of the mortality at various stages of *A. mali*. The parasitic wasp *Atanycolus denigrator* (Hymenoptera: Braconidae) and parasitic mite *Pyemotes* sp. (Acariformes: Pyemotidae) are the dominant natural enemies. Chemical control currently is the major management strategy in infested orchards in China, which

often involves applications of systematic and contact insecticides to infested trees. In addition, removal of insect-infested branches can be effective against the dispersal of *A. mali* populations, and quarantine measures have been taken to circumvent human-assisted spread of this pest. A biological control program has been recently carried out by releasing the parasitoid *Sclerodermus pupariae*. More than 5 million parasitoid wasps have been released since 2009; their efficacy in controlling *A. mali* is still being evaluated.

Emerald Ash Borer Risk Assessment and Sampling Design

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ABSTRACT

The use of statistical models to determine nationwide sampling strategies can maximize the efficient use of sampling resources. Such models can also provide an objective and transparent process for the selection of trap locations. This methodology has been applied to the sampling efforts of emerald ash borer (EAB) (*Agrilus planipennis*), where detection likelihood was modeled using the Maximum Entropy (MaxEnt) statistical technique. This was done with specific focus on increasing the number of successful detections beyond the known infested area. MaxEnt identifies significant associations between EAB presence data with selected site variables and then maps the probability of EAB presence in a geographic context. Two separate detection likelihood models were developed: 1) the “all points” model which used all known EAB positive locations from 2002 to 2013 and 2) the “2013 only” model which used only positive locations reported in 2013, thereby creating extra emphasis on site characteristics associated with the frontier of the EAB range, which may be different from characteristics in the known infested area. A suite of independent variables representing geographic, ecological, and human demographic characteristics were used in the detection likelihood models. The MaxEnt bootstrap option was used to spatially quantify model uncertainty. Suggested trap locations for the 2014 EAB field season were generated using the Forest Health Technology Enterprise Team sample design tool. Potential sample areas were further restricted to locations that intersected roads and were likely to contain ash species. For the 2014 EAB sample design, 22,000 primary sample locations were generated along with an additional 4,400 replacement locations provided as alternatives. Each sample location represents a 1 km² sample unit within which EAB detection and monitoring efforts are recommended. This sampling design has demonstrated success with sampling of EAB and shows similar promise for a more effective, objective, and transparent process for other sampling efforts.

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