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INCOMPATIBILITY BETWEEN TWO SUB-SPECIES OF *LYMANTRIA DISPAR* (LEPIDOPTERA: LYMANTRIIDAE)

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ABSTRACT

Knowledge of the compatibility of gypsy moth, *Lymantria dispar*, strains from other world areas with the strain present in North America has become important since the recent introductions of gypsy moths from Eurasia. Incompatibilities, if found, could result in infertility of hybrids and reduce the chance of the newly introduced genes spreading within the gypsy moth population. Previously, we evaluated the compatibility of several geographically separate Eurasian strains (two European and four Asian) with the North America gypsy moth. There has been no indication of incompatibility in the F_1, F_2, or backcrosses produced from crosses between any of these Eurasian strains and the North American strain. All of the gypsy moth strains assessed so far were of the same sub-species, *L. dispar dispar*. Recently, we crossed the *L. dispar dispar* subspecies (Rocky Hill, Connecticut) and the *L. dispar japonica* subspecies (Nagoya, Honshu Island, Japan) to assess compatibility. *Japonica* adults of both sexes are larger than those of *dispar*. The base color of the *japonica* female's wings is gray or brownish with less distinct black markings than *dispar*. The wings of the *japonica* males are a chocolate to dark brown, similar to the darker color forms of *dispar*.

Hatch of the F_1 hybrids was consistent within each of the females' parent strains, indicating no incompatibility in the egg stage. Additionally, there was no evidence of increased mortality in the hybrids due to incompatibility between the subspecies. However, there were differences in developmental rates that could temporally isolate the two subspecies if they coexist in nature. The *dispar* larvae developed significantly faster than the *japonica* larvae, while the F_1 hybrids had developmental rates intermediate to the parentals. The differences in developmental rates of the *dispar* and *japonica* larvae appear to be due both to rate of growth and number of instars.

Forty-seven percent of the adult females resulting from the *dispar* x *japonica* (mother x father) cross had abnormal morphology and were infertile, indicating incompatibility between these two subspecies of *L. dispar* in the F_1 generation. All of the other F_1 adults (both sexes) had normal morphology and were fertile. Abnormal females were indistinguishable from normal females as pupae, but had more difficulty in eclosing and expanding their wings. Abnormal females had female shaped wings, male wing coloration, and tended to be smaller in size than normal females. In addition, the antennal branches in the abnormal females were intermediate in length

between that of normal males and females. The abnormal females attempted to mate but never produced an egg mass, even though eggs were present in their abdomens. Based on this evidence, we conclude that the *dispar* and *japonica* subspecies of *L. dispar* are incompatible and may actually be distinct species.

PROACTIVE RESEARCH ON *LYMANTRIA MONACHA* (LEPIDOPTERA: LYMANTRIIDAE) TO PREVENT ITS INTRODUCTION AND ESTABLISHMENT

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ABSTRACT

Lymantria monacha (nun moth), not known to be established in North America, is an Eurasian pest of conifers (spruce, fir, larch and pine) that poses an ever-present threat of being accidentally introduced because of its biology and behavior. Its establishment would be disastrous because of its polyphagous feeding habits, ability to colonize new habitats, and capacity to be spread rapidly by vagile adults. Adults are readily attracted to artificial lights and have been observed in Russian Far East ports. Nun moth has a high potential to be transported via commerce because, although eggs are normally laid in tree back crevices they also could be deposited in crevices on containers, pallets, ships, etc. To develop tactics to prevent its introduction and establishment in North America, initially we are studying its biology, developing techniques to identify each life stage, and differentiating it from *Lymantria dispar*, a closely related species.

Biology. The life history of the nun moth is similar to that of the gypsy moth (*L. dispar*). Adult nun moths fly from the middle of July to the beginning of September. Nun moth adults are most active after midnight; males are much more active than females. Once mated, females lay 70 to 300 eggs in separate clusters of approximately 40 eggs bearing no hair covering. After almost completing its development, the embryo goes through a winter diapause before hatching, which usually occurs in the beginning of May. First and second instars are capable of being wind-dispersed for considerable distances. Larvae go through 5 to 7 instars before they pupate in July.

Identification. Mature nun moth larvae, 30 to 40 mm in length, are tan, green or dark-gray in color, with extensive brown or black mottling. Each dorsal verruca on the 3rd thoracic segment of the hairy larva is nearly encircled by a separated white patch. The larvae usually also have a light patch that fills the middorsal space between the verrucae from the middle of the 4th to the middle of the 6th abdominal segments. The dorsal glands in the middle of the 6th and 7th abdominal segments are prominent and orange in color. Beginning with the 3rd instar, the larval head is orangish-brown with numerous brown and black freckles. The pupa has no cocoon, is reddish brown and shiny, with light colored clumps of hair. In adults of both sexes, the coloration of the forewing can vary from the characteristic chalk-white, decorated with numerous dark transverse wavy lines and patches, to almost black. The hind wings are generally gray-brown with minute dark and/or light patches at the edge. The female has a wingspan of 45 to 55 mm, and the male has a wingspan of 35 to 45 mm. The female has a broader body than the male and an extremely long ovipositor adapted for its specialized egg-laying habit.

PEST RISK ASSOCIATED WITH THE IMPORTATION OF UNPROCESSED WOOD

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ABSTRACT

The risk of exotic pests entering the United States on unprocessed wood is an increasing concern. Past introductions of forest pests, such as the Chestnut Blight, Dutch Elm Disease, and Gypsy Moth, have significantly changed the ecology within North American forests. The recent introduction of the Pine Shoot Beetle (*Tomicus piniperda*) and Asian Long-horned Beetle (*Anoplophora glabripennis*), with the associated costs in management and eradication of these pests, reemphasizes the importance of preventing the introduction of additional forest pests.

The importation of unprocessed wood products presents a risk of importing associated pests. Unprocessed wood enters the United States either as "logs and lumber" or as "solid wood packing material." The importation of logs and lumber requires a permit. The conditions stated in the permit must be followed to prevent the risk of importing pests. Common requirements for logs and lumber include kiln drying, fumigation or be destined to an approved facility. At entry into the United States, inspectors verify the import's suitability.

Solid wood packing material does not require a permit to enter the United States. Solid wood packing material includes lumber used for dunnage, crating, skids and pallets associated with a cargo. Solid wood packing material presents a risk, because it often arrives untreated and may harbor wood pests. Large amounts of solid wood packing material are entering the United States. For example, more than 2.5 million containers entered through the Ports of Seattle and Tacoma in 1997. Heavy cargo, such as ironware and stone, uses large dimension lumber to hold the cargo in place during shipment, but virtually any shipment can contain unprocessed wood. Shipments are prioritized for inspection, and if they contain infested solid wood packing or if bark is present, the shipment must be treated to prevent any pest risk.

Forest insect pests are often intercepted with these wood products. Scolytidae is by far the most common group of insects. Scolytidae interceptions include: *Pityogenes calcographus*, *Ips erosus*, *Hylurgops palliatus*, and *I. typographus*. Other common insect interceptions include: *Pissodes sp.*, Curculionidae; *Monochamus sp.*, *Xylotrechus sp.*, Cerambycidae. Until recently most interceptions occurred on the east coast, but more interceptions are coming with cargo on the west coast as markets expand on the Pacific Rim.

GYPSY MOTH TECHNICAL INFORMATION PROJECT: PAST AND PRESENT

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ABSTRACT

The Gypsy Moth Technical Information Project (GMTIP) was conceived in 1975. Its purpose was to make the worldwide technical literature on the gypsy moth available to participants in the USDA Expanded Gypsy Moth Research and Development Program. The start-up cost was $555,000. Specific objectives included assembly of documents pertaining to the gypsy moth and related topics, preparing a computer database of these documents, and making this available to users. By 1979, more than 4,000 documents had been collected. The database included the title, author, year, source and an abstract, as well as keywords, geographic locations, taxons, language, location of the original document, and the National Agricultural Library call number for each document. The titles, abstracts, and for some articles the full text were translated for more than 600 foreign language documents. Microfiche copies were made for about 3,600 documents. Documents were added until 1986.

Preparation of the electronic (computer) information database was an intensive 3-year project. Collection of documents and additional bibliographic information was done at Pennsylvania State University. Computer entry was done at Data Courier, Louisville, Kentucky. The database was created and maintained by the Oak Ridge Computerized Hierarchial Information System (ORCHIS). This program and the computers at Oak Ridge also were used by the Forest Service for its Forestry Technical Information System. The FTIS was later transferred to Fort Collins, Colorado. Unfortunately, copies of the GMTIP computer tapes cannot be located. The last known use of the GMTIP database is a printout made in 1984. (by WCC is the only identification of where the print out was made).

Facsimile copies of the GMTIP source documents still exist. During the 1980's and early 1990's, photocopies of the original documents resided with State and Private Forestry in Morgantown, West Virginia, and the Carnegie Museum in Pittsburgh, Pennsylvania. In 1997, the remaining physical materials of GMTIP (photo- and microfiche copies of documents, author card files, printouts of the entire database and the taxon and keyword indices) were transferred to the NERS Laboratory at Ansonia, Connecticut. The GMTIP source documents are arranged numerically in file cabinets and are readily accessible. There are at least 4,650 documents at Ansonia. Originals of most of these documents can be found in the National Agricultural Library.

Acknowledgment is given to Robert Acciavatti for preparing card files and caring for the documents for many years, to Sandy Liebhold for helping to bring the documents to Ansonia, Connecticut, and to Harry Hubbard for organizing the documents into file cabinets.

CHINESE COCCINELLIDAE FOR BIOLOGICAL CONTROL OF THE HEMLOCK WOOLLY ADELGID

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ABSTRACT

The hemlock woolly adelgid, *Adelges tsugae* Annand, is an introduced pest that is causing mortality of hemlock in the Eastern United States. The adelgid is native to Asia and occurs in Japan, China, and India. It is not considered a pest in Asia where host resistance and natural enemies combine to keep populations at innocuous levels. It has no effective natural enemies in the United States.

We surveyed for natural enemies of hemlock woolly adelgid (HWA) in Sichuan and Yunnan Provinces, People's Republic of China. The adelgid and associated natural enemies were found on the Chinese Asian hemlocks *Tsuga dumosa* and *T. forrestii*, which grow in the mountains between 2300 and 2900 meters. In China, the adelgid is attacked by a complex of predators in eight families in four orders: Anthocoridae (Hemiptera); Cecidomyiidae, Chamaemyiidae and Syrphidae (Diptera); Chrysopidae and Hemerobiidae (Neuroptera); Derodontidae and Coccinellidae (Coleoptera). The family Adelgidae has no known predators or diseases and none were found in China. The diversity of Coccinellidae found on HWA-infested hemlock is extraordinary. To date, more than 53 species have been collected, 31 of which appear to be species that have not been described previously. Most of the new species are in the tribe Scymnini.

Three newly described species have been imported to the USDA Forest Service Quarantine Laboratory at Ansonia, Connecticut. One species, *Scymnus sinuanodulus* Yu et Yao 1997, has been reared through a complete generation in the laboratory. It lays eggs in the spring and appears to be univoltine. The eggs hatch in about a week and larvae complete development in about 4 weeks. The adults become dormant at low temperatures, but do not enter diapause. Our host range evaluations of this species indicate that it prefers adelgids but will feed on aphids if starved. We believe that *S. sinuanodulus* is a promising candidate to release and establish for biological control of the HWA.

DENDROCHRONOLOGY OF GYPSY MOTH OUTBREAKS IN THE NORTHEAST

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ABSTRACT

Using data from the intensive plot system (IPS) study, we examined the tree ring chronologies from several thousand trees, both gypsy moth hosts and non hosts. The data were collected from five areas in the Northeastern United States, in the generally infested area. We found that moderate to severe gypsy moth outbreaks may be detected as declines in average oak (*Quercus*, spp.) increment and that at least one historical outbreak was evident in chronologies from western Massachusetts. Using step wise regression, we found that increment of host species was negatively affected by defoliation in both the individual tree and area average increment data. The effect of defoliation on increment appears to be greatest in the same year as defoliation but there may also be a decline in growth in the year following defoliation. Surprisingly, aspen (*Populus grandidentata* and *P. tremuloides*) appears to respond to defoliation with less magnitude than oaks despite high levels of defoliation. Compensatory increases in growth increment may be observed in some, but not all, non-host species during outbreaks. Ash (*Fraxinus*, spp.) and yellow-poplar (*Liriodendron tulipifera*) increased growth in response to stand level gypsy moth defoliation.

MICROSPORIDIA: A POTENTIAL AGENT FOR BIOLOGICAL CONTROL OF GYPSY MOTH (*LYMANTRIA DISPAR* L.)

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ABSTRACT

Experimental plots were established in three different regions of the Slovak Republic: Čifáre, Rimavská Sobota, and Trebišov in March, 1996. In 1997, an additional plot was established in the locality of Bušince. Burlap bands were fixed on 100 oak trees in each plot for the purpose of collecting late-instar gypsy moth larvae. Larvae were collected from each plot in three different stages: L1-2, L3-4, and L5-6. Early instar larvae were collected using the foliage-beating sampling method.

All larvae that were collected from each plot were either dissected individually or homogenized in a blender as a water suspension. Thirty to fifty larvae were dissected individually whereas excess larvae were homogenized as a group. Light microscopy was used to detect the presence of microsporidian spores in larval tissues or in the homogenate. Suspension of spores were treated with streptomycin sulfate to control bacterial contamination and sent to scientists at the Illinois Natural History Survey (INHS) for identification and bioassay.

Microsporidia from the genera *Vairimorpha* and *Nosema* were recovered from the plots but at low levels of prevalence. This was not unexpected because the densities of gypsy moth larvae were very low in all regions of Slovakia in 1996-97. Usually only one genus of microsporidia was common to each site. Individual larvae frequently contained mixed infections containing microsporidia, NPV, fungi, and bacteria, though the overall level of mortality was low.

The *Vairimorpha* isolate (probably *Lymantriae*) was propagated in the laboratory at the INHS, prepared as a clean inoculum, and sprayed on the foliage of young oak trees in a 50 X 50m plot at a dose of 1 X 7^{10} spores in a water suspension. All visible larvae were collected from the inner portion of the plot (40 X 40m) 14 days after treatment and dissected individually for infection. From this sample, ca. 50% of the larvae were infected, which was encouraging. We will continue to measure the prevalence of infection in the experimental plots and evaluate the feasibility of using microsporidia for augmentative biological control of gypsy moth populations in the Slovak Republic and elsewhere.

EXAMINING THE ROLE OF DELAYED-INDUCED RESISTANCE IN GYPSY MOTH OUTBREAKS

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ABSTRACT

We tested the effects of fertilization and gypsy moth defoliation on the expression of delayed-induced resistance in a large scale, controlled field experiment. Defoliated, undefoliated, N-fertilized, and unfertilized treatments were replicated 4 times in 1/4 ha blocks of Populus x euramericana. More than 2 million gypsy moth neonates were released in 1996 and an additional 10 million in 1997 to create outbreak population densities in the defoliation plots. Undefoliated control plots were kept free of gypsy moth with a system of tangletrap barriers. Defoliation significantly increased foliar concentrations of total phenolics and condensed tannins in both 1996 and 1997. Fertilizer mitigated the induction of these secondary chemicals in 1996, congruent with predictions of the carbon-nutrient balance hypothesis. However, fertilizer had no effect on secondary chemistry in the second year of defoliation. Phenolic concentrations were positively correlated with gypsy moth density, marking perhaps the first time that the chemical composition of a forest can be related to the population density of an insect. No effect of foliar chemistry on gypsy moth performance was apparent in 1996, the first year of defoliation. In 1997, female pupal mass was negatively correlated with both phenolic concentration and gypsy moth density in the plots. These data are consistent with the hypothesis that delayed-induced resistance can mediate a delayed density-dependent decrease in growth rates of gypsy moth populations.

MICROSPORIDIA FROM GYPSY MOTH POPULATIONS IN BULGARIA

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ABSTRACT

Several species of microsporidia occur in the gypsy moth populations in Europe. They are considered to be important components of the natural enemy complex of this forest defoliator. Mirchev *et al.* presented data about microsporidian infections in low dense population in the Northeastern part of Bulgaria, but no prevalence data have been published.

We documented microsporidian infections and monitored their prevalence in several gypsy moth populations in Bulgaria. Microsporidia have been found in the gypsy moth populations in Southwestern Bulgaria - Rupite, Northwestern Bulgaria - Levishte and Central Bulgaria - Asenovgrad. One species occurred in each of these populations, *Vairimorpha* sp. in the Rupite area, *Nosema* sp. in Levishte and *Endoreticulatus* sp. in Asenovgrad. The prevalence of *Vairimorpha* sp. was monitored from 1984 to 1997. It fluctuated during this period even when gypsy moth densities were low. The prevalence of *Nosema* sp. was monitored from 1996 to 1997. A decline of the percentage of the infected larvae was recorded in 1997. It may be due to the drastic decrease of gypsy moth population density probably initiated by the spraying of Bt and chemical insecticides. The prevalence of *Endoreticulatus* sp. was monitored in 1996 and 1997. It was found that it increases within the age of the gypsy moth larvae both in 1996 and 1997.

The three microsporidian species found in Bulgarian gypsy moth populations may be important in the regulation of population densities. Long-term monitoring could establish a fairly complete picture of the effects of these microsporidia on gypsy moth populations and influence possible decisions made regarding microbial or chemical insecticides.

UPDATE ON THE GENOTYPE ANALYSES OF SPECIMENS

TRAPPED IN THE 1997 ASIAN GYPSY MOTH PORT SURVEY

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ABSTRACT

Since 1994, high-risk ports in the U.S. have been surveyed for the presence of the Asian gypsy moth. These include ports both inside and outside the generally-infested area which is concentrated in the northeast U.S.. The Port Survey was initiated as a result of introductions into Washington and Oregon (1991), North Carolina (1993), and potentially several east coast ports (1994). The Asian strain is considered a more serious pest than that found in North America since its larvae have a broader host range and adult females can fly long distances. Females of the existing North American strain are unable to perform active flight. Specimens trapped in several states (15) have been submitted for genotype analyses because no morphological characters exist that can be used to differentiate the two strains. Because of the large number of specimens submitted (~ 20,000), they are prioritized since the analyses severely limits throughput. Four genetic markers (mtDNA, FS1, 10F1, and 9C2) are being used in the analyses, which includes the following experimental steps for each *individual* specimen: 1. Removal from the trap (Delta or milk carton), 2. Tissue extraction, 3. Loci amplification via PCR, 4. Restriction enzyme digestion if necessary, and 5. DNA fragment analysis via electrophoresis. Of the 1529 specimens analyzed as of 10 January 1998, 'Asian' gypsy moths have appeared only at Long Beach, California and Seattle and Tacoma, Washington. These results are compared to those of Port Surveys of previous years.

INVASIVE TREE SEED PESTS: CONDITIONS FOR SUCCESSFUL ESTABLISHMENT

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ABSTRACT

Worldwide exchange and trade of tree seeds is rapidly increasing with the development of plantations and ornaments using exotic tree species. Spermatophagous insects (i.e., entirely developing within seeds), thus constitute serious potential invaders in most countries. A total of 58 spermatophages are known to attack conifer seed cones. Most of them belong to the seed chalcid genus *Megastigmus* (Hymenoptera: Torymidae). A 20-year (1977- 1997) survey of *Megastigmus* damage has been carried out in Europe, Asia Minor, and north Africa on 115 species of trees and shrubs including both native and exotic species. A total of 56 conifers (29 native, 37 introduced to Europe), 24 Rosaceae, and 5 Anacardiaceae were infested by 20 species of *Megastigmus*. Among these species, 13 were native of Europe and Asia Minor, and 7 were introduced from north America.

Some chalcid biological patterns were shown to help in the colonization process; i.e., capability of developing in unfertilized seeds, parthenogenetic (either thelitokous or arrhenotokous) development, and prolonged diapause. However, the establishment and spread of exotic seed pests in the area of introduction essentially depends on the presence of native tree species congeneric to the original host. When such congeners exist, the introduced chalcids are observed to shift on most of them. Four species attacking firs in the US (*M. milleri*, *M. pinus*, *M. rafni*, and *M. specularis*) thus attack most of the European and Caucasian firs in Europe and Asia Minor. Because chalcids are the latest species to attack, competition with native cone insects however limits the percentage of damaged seeds in most cases. The relative length of female ovopositor with regard to the cone size may also limit the host range. When there is no native tree congeneric to the original host, the introduced chalcid cannot develop. However, when the original host is simultaneously introduced, the chalcid tends to occupy entirely the seed cone niche because of both the absence of native and introduced competitors and a limited parasitism.In such case, seed damage is very important. The Douglas-fir seed chalcid, *M. spermotrophus*, can attack up to 100% of seeds in European seed orchards.

Because Mediterranean cypress began to be introduced from Greece towards the western Mediterranean more than 2000 years ago, the relationships between cypress and the specific seed chalcid, *M. wachtli*, were interesting to be precised. Analysis of DNA microsatellites (7 loci studied) in chalcid populations from the whole Mediterranean range revealed three separate groups: i) Greek populations from natural stands, showing a high variability but

populations of Crete differed from these of islands of Eastern Aegean Sea; ii) populations from northern Greece; iii) populations from France, Italy, Tunisia, and Algeria, characterized by a high homozygoty (bottleneck effect), that appear closer to Cretan populations than to these of other natural areas. The long-term and continuous introduction of cypress in continental Greece thus results in a higher diversity of chalcids than in the western Mediterranean.In conclusion, insect survey must be reinforced at introduction of tree seeds with no present congeners in Europe (e.g., *Chamaecyparis*, *Cryptomeria*, *Thuja*).

DEVELOPMENTAL MODELS FOR THE TACHINID PARASITOIDS *BLEPHARIPA PRATENSIS* AND *PARASETIGENA SILVESTRIS* IN *LYMANTRIA DISPAR*

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ABSTRACT

Insect life system models typically use temperature-dependent functions, either as a linear regression of development to temperature or as a boundary layer problem with matched asymptotic expansions, as solutions. Temperature-independent models with variability also can be built using a stochastic approach based on "Same Shape Property" of normalized cumulative frequency distributions fit to a cumulative Weibull function. Larval development in the tachinids *Blepharipa pratensis* (Meigen) and *Parasetigena silvestris* (Robineau-Desvoidy), introduced to North America as biological controls of gypsy moth, *Lymantria dispar* L., has been described previously using means and standard deviations of development time. I estimated the larval development for both *B. pratensis* and *P. silvestris* and used those estimates in developing and comparing approaches to modeling the influence of temperature on their development.

The parasitoids were field collected in Connecticut, and reared on host larvae from a laboratory strain of *L. dispar* grown at the Northeastern Center for Forest Health Research, Hamden, Connecticut. For each tachinid, 4th and 5th instar larvae were parasitized and reared in environmental chambers set at 15°C, 20°C, 25°C, and 30°C. The days from parasitization to larval emergence, and the reciprocal values were used to estimate the developmental parameters. Models were derived using a linear regression, the boundary layer method, and the "Same Shape" approach.

The linear regression approach provided the closest association between observed and predicted values. The slopes of the linear equations for tachinid larval development were higher in the host 5th instar. Intercepts of the linear equations were significantly different for each tachinid regardless of host instar. Frequency histograms of larval development in both tachinids broadened with increasing temperature and were relatively asymmetric toward longer development times. The temperature-dependent models had similar shapes across the range of temperatures, though the combined models lacked good fit at the higher temperatures.

Along the range of temperatures to which a parasitoid is adapted, the linear regression approach seems adequate for modeling its development, but may become less adequate as temperature nears or exceeds lower or upper optima. The temperature-independent modeling approach, by

including inherent variability, may better simulate population changes under more variable conditions of environmental temperature. Temperature affects on larval development can be immediate in parasitoids like *P. silvestris* that begin feeding immediately in hosts, or delayed as in species like *B. pratensis* where feeding is delayed until hosts reach the pupal stage. The selection of an approach for modeling populations ultimately remains linked with use of the model, but approaches that include variable aspects of development may be most appropriate.

GENETICS OF ALLOZYMES IN FLIGHT- AND NONFLIGHT-CAPABLE FEMALES IN LABORATORY STRAINS OF *LYMANTRIA DISPAR*

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ABSTRACT

Female gypsy moth, *Lymantria dispar* L., flight is a character that follows a broad clinal pattern in their Palearctic range, starting with no flight in the West and increasing to full flight in the East. According to Eurasian researchers, gypsy moth larvae indigenous to eastern Eurasia and Japan may also have a broader host preference for conifers. With increased global trade and movement, Far Eastern strains have inadvertently been introduced into southeastern and northwestern North America. Analyses of genetic variability in gypsy moth have used structural protein loci to identify the probable geographic source of a population. The loci also represent large-scale genetic markers that could be associated with other polygenic traits like flying capability. I obtained estimates of genetic variation in laboratory strains of gypsy moth and compared them to estimates of female flight capability determined by other researchers.

The genetic variability in four laboratory strains of *L. dispar* was evaluated at eight enzyme loci using horizontal starch gel electrophoresis. The strains originated from eggs collected in Germany (GL), Far Eastern Russia (RM), Siberian Russia (RBI), and Connecticut (CT) in USA. The measures of heterozygosity were used to estimate genetic diversity. The findings from other researchers (Keena, Wallner, and Grinberg, personal communication) that female flight capability estimates were least in the CT population, intermediate in GL and RM, and highest in the RBI samples, were used for comparison with heterozygosity. Similarly, genetic diversity was found to be highest in the Siberian population at two of the loci. The CT and RM populations each had one locus with two alleles, while the GL population was heterozygous for a different locus. The graphical representation of genetic heterozygosity versus female flight capability suggests this behavioral character may be associated with neutral allozyme markers. An unpaired group mean weighted analysis using Nei distance measures also affirms the increasing genetic diversity of gypsy moth from the eastern Palearctic, with the Eurasian populations all being clumped.

Classic hybridization studies by Goldschmidt and others, and recent allozyme studies in other laboratories, suggest that Far Eastern population may be distinct subspecies. With the exception of Goldschmidt, none of the other studies included estimates of female flight capability. Further research will include assays with a population of *L. dispar* from Japan. Studying populations of gypsy moth from a broader geographic range may increase the number of polymorphic loci with which to investigate the genetic character of female flight capability.

MEASURING AND MANAGING THE RATE OF GYPSY MOTH SPREAD

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ABSTRACT

The Slow-the-Spread (STS) pilot project was initiated by the USDA Forest Service in 1993 to determine the feasibility of slowing the spread of the gypsy moth in specific portions of North Carolina, Virginia, West Virginia and Michigan (Leonard and Sharov 1995). To evaluate the effect of STS activities on the rate of population spread we suggested to use 2 criteria: (1) population spread rate = the distance between population boundaries in consecutive years, and (2) inter-boundary distance = the distance between population boundaries estimated in the same year using different population thresholds. Both criteria indicated that the rate of population spread in the Appalachian Mountains declined after 1990 when suppression of isolated infestations started within the Appalachian IPM project and then continued within the STS project. In 1996-97 gypsy moth populations declined in Virginia and West Virginia apparently because of the fungal pathogen *Entomophaga maimaiga*, and the rates of spread became negative. We developed a model of population expansion which assumes that the probability of establishment of new colonies decreases with increasing distance from the population front and the number of individuals in each colony grows exponentially (Sharov and Liebhold 1998). The effect of barrier zones was simulated by truncating the function of the colony establishment rate to zero beyond a specific distance from the population front. This model predicted a 54% reduction of the rate of gypsy moth spread in the Appalachian Mountains, which is close to the actual 59% reduction. Possible expansion of the STS project to a national scale requires understanding the differences in the pattern of population spread among various geographic areas. In Michigan, the rate of gypsy moth spread did not depend on winter temperatures but was positively correlated with habitat quality (Sharov *et al.* 1998). The rate of population increase and dispersal rates in various states were estimated from the rates of spread and inter-boundary distances. In the Appalachian Mountains, gypsy moth populations had a limited dispersal but a high rate of population increase. In eastern Virginia (coastal plain), gypsy moth populations had a high dispersal rate but a low rate of population increase. And in Wisconsin, both the dispersal rate and the rate of population increase were high, causing a fast progression of the population front.

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HEMLOCK WOOLLY ADELGID: A RESEARCH UPDATE

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ABSTRACT

The hemlock woolly adelgid (HWA), *Adelges tsugae* Annand, is an exotic pest native to Asia. It was first discovered in North America (Oregon and British Columbia) in the early 1920s. It was found on the East Coast in Virginia in 1951. The HWA now occurs from North Carolina to the northern border of Massachusetts where it has caused extensive damage to eastern hemlock, *Tsuga canadensis* (L.) and Carolina hemlock, *T. caroliniana* Engelm. Our research objectives are to understand the biology of this exotic insect and its interactions with host plants and environmental stressors, and to develop ecologically sound strategies for management of HWA. Progress is as follows:

<u>Host Interactions.</u> We have observed that adults and nymphs of all generations of HWA insert their feeding stylets into the tissue at the base of hemlock needles (the leaf cushion) and feed on the parenchyma cells of the xylem rays, secreting considerable amounts of saliva in the process. By feeding on the contents of storage cells, the HWA depletes the tree's resources and may render it more susceptible to other environmental stressors.

<u>Impacts.</u> We used springtime satellite images and a variety of vegetation indices to identify and classify the health of hemlock forests within a 428-mi^2 HWA-infested area in the lower Connecticut River Basin. Field data, based on the U.S. Forest Service Crown Condition Rating Guide, were collected at 150 sites within the study area and were used to verify and refine the health classifications derived from the satellite images. Our health classification technique was then applied to 1985, 1988, 1993, and 1995 satellite images of the study area so that trends in the health of hemlock forests could be identified.

Overall, there was a modest decline in hemlock health since HWA was first discovered in Connecticut in 1985. Hemlock health declined dramatically between 1988 and 1993, but improved somewhat in 1995 in most locations within the study area. An analysis of available data from the study area indicates that site characteristics are related to hemlock decline. There was less decline in the health of hemlocks on northwest-, north-, northeast-, and east-

facing slopes than in those on southwest- and west-facing slopes. Hemlocks on ridge tops and upper slopes showed a greater decline in health than those in adjacent valleys or riparian zones.

Biological Control. Predators in the families Coccinellidae, Chamaemyiidae, Cecidomyiidae, Chrysopidae, and Inocelliae have been found feeding on HWA in China. To date, more than 50 species of Coccinellidae have been found, at least 30 of which are new to science. Three of the newly described species have been imported into the USDA Forest Service Quarantine Laboratory in Ansonia, Connecticut. One species, *Scymnus sinuanodulus* Yu et Yao, has been reared through a complete generation in the laboratory and its host range has been evaluated. It is a very promising candidate for biological control; experimental releases are planned for 1998.

LABORATORY AND FIELD STUDIES USED TO EVALUATE THE HOST SPECIFICITY OF GYPSY MOTH MICROSPORIDIA

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ABSTRACT

Host specificity of a biological control agent is a difficult characteristic to measure prior to release of the agent because data generated in a laboratory setting may not accurately predict host range in the field. The laboratory (physiological) host range of most insect pathogens is generally considered to be far broader than the ecological host range. We performed classical host specificity testing of microsporidian biotypes from European *Lymantria dispar* populations against native nontarget lepidopteran species and found that 50% or more of the nontarget species were susceptible. Not all infections, however, were comparable to infection in *L. dispar*, the natural host. Using *L. dispar* as a model nontarget host with a known field history (no microsporidia in North American populations), we fed viable spores of microsporidia isolated from nine species of sympatric forest Lepidoptera sympatric to the *L. dispar* larvae. All of the microsporidia infected *L. dispar*. These data indicate that a host infected by a microsporidium under laboratory conditions may not be in the ecological host range of the microsporidium.

In an effort to determine the level of ecological complexity at which the microsporidia exhibit host specificity, we placed infected nontarget host larvae (*L. dispar*) with uninfected *L. dispar* larvae in confined laboratory arenas. Three of the nine microsporidian species were transmitted from infected to uninfected larvae, although at lower rates than rates of transmission between the natural hosts of these species. When infected and uninfected larvae were placed on sleeved host plant foliage, a more heterogeneous arena, no transmission occurred. Thus, simple but ecologically more complex bioassay experiments may be useful in predicting ecological host specificity.

We are evaluating our predictions of ecological host range by studying the host specificity of the *L. dispar* microsporidia in the aboriginal range. We have surveyed the pathogens of nontarget lepidopteran populations from areas in Bulgaria where three microsporidian genera are endemic in *L. dispar* populations. Four microsporidian isolates were found in tortricid and noctuid hosts which were feeding on the same host plants with *L. dispar* larvae. Preliminary evaluations indicate that the microsporidia are not *L. dispar* pathogens.

COMPARISON OF DIVERSITY OF NONTARGET ARTHROPODS BETWEEN SIMILAR OAK-DOMINATED APPALACHIAN FORESTS PRIOR TO BIOLOGICAL TREATMENT OF GYPSY MOTH

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ABSTRACT

In 1994, 18 200 ha plots, nine each on the George Washington and Monongahela National Forests, were designated for research on impact of biological insecticides for gypsy moth (*Lymantria dispar* (L.)) suppression on nontarget forest arthropods, songbirds and salamanders. Baseline sampling for arthropod diversity was carried out in 1995 and 1996 by means of blacklight traps, Malaise traps, pitfall traps, canvas tree bands and foliage pruning.

Distribution and host associations are being studied for the following number of species/taxa identified from the baseline year samples: Macrolepidoptera (492 species), Formicidae (29 species), Symphyta (14 species), parasitoids reared from Lepidoptera (38 Tachinidae species, 35 Hymenoptera species), Pentatomidae (11 species), Carabidae (68 species), Chrysomelidae (25 species), Curculionidae (21 species), Tachinidae (108 species) and Araneae (311 species). Herbivorous taxa are being evaluated on red maple, hickories and oaks.

For Macrolepidoptera, Carabidae and Araneae, species richness and abundance over the season have been determined. Species accumulation curves have been developed and approach zero at the end of the second season indicating that existing fauna are well represented in samples. While total faunal richness for Lepidoptera, Carabidae and Araneae is similar between the two forests, species composition is different. Faunal similarity between the two forests is 75% for Macrolepidoptera, 62% for Carabidae and 61% for Araneae.

EFFICACY OF GROUND-BASED APPLICATIONS OF GYPCHEK AND AN *IN-VITRO* PRODUCED NPV

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ABSTRACT

At present, high production costs dissuade commercial interests from developing and marketing Gypchek, the gypsy moth nucleopolyhedrosis virus (LdNPV) product registered with the U.S. Environmental Protection Agency as a general use pesticide for aerial and ground application to control the gypsy moth. However, a recent survey of gypsy moth managers, including commercial arborists, revealed a high level of support for research and development to improve Gypchek through discovery of more effective viral strains and formulations (Podgwaite *et al.* 1997, USDA Forest Service, Northeastern Forest Experiment Station, Gen. Tech. Rep. NE-240.). Recently an LdNPV strain (122b1a) with great potential for scaled-up *in-vitro* production, and eventual replacement of the current *in-vivo* product, has been developed (Slavicek, J.M. and M.J. Mercer, 1995, U.S. patent #5,420,031). Also, Gypchek formulation evaluations by USDA scientists have shown the stilbene, Blankophor BBH (Burlington Chemical, Burlington, NC) to be an enhancer of LdNPV activity and, potentially, a cost-reducing formulation adjuvant (Webb *et al.* 1996, J. Econ. Entomol. 89: 957-962).

In 1996, we evaluated ground-based applications of various doses of Gypchek with and without sunscreen and viral enhancer and compared them to a *Bacillus thuringiensis* treatment. In 1997, a formulation of the *in-vitro* produced 122b1a and a reduced dose of Gypchek were evaluated in addition to the non-enhancer treatments tested in 1996. Treatments were applied to overstory oaks in the Glassboro Wildlife Management Area, Glassboro, NJ, and evaluated on the basis of larval mortality, live larval abundance, foliage protection, and change in egg mass density.

Results in 1996 indicated that Gypchek at a concentration of 10^{10} polyhedral inclusion bodies (PIB) per gallon in a formulation that contained the sunscreen Lignosite AN (Georgia Pacific,

Bellingham, WA) was no more efficacious than either the same dose without the sunscreen or a dose of 10^9 PIB per gallon in a formulation that contained both the sunscreen and Blankophor BBH. No Gypchek treatment was as effective as *Bacillus thuringiensis* (Foray 48B, Abbott Laboratories, Chicago, IL) in reducing larval populations but all Gypchek treatments were as effective as Foray 48B in protecting foliage.

In 1997, a reduced dose of Gypchek at $5X10^9$ PIB per gallon of formulation was as effective as 2-fold higher doses in reducing larval populations and protecting foliage. However, the *in-vitro* produced 122b1a formulated at 10^9 PIB per gallon was significantly less effective in reducing larval populations than the same dose of Gypchek. Subsequent studies have revealed that 122b1a PIBs are significantly smaller than Gypchek PIBs and contain fewer virions. That may account for the differences in results seen in this study. Further studies will be necessary to establish dose and formulation parameters for the *in-vitro* produced 122b1a.

THE IMPORTANCE OF THE EIGHT-TOOTHED SPRUCE BARK BEETLE

(*IPS TYPOGRAPHUS* L.) IN CENTRAL EUROPE

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ABSTRACT

Ips typographus is the most serious bark beetle pest of mature spruce stands in Central Europe. This pest is responsible for the loss of from 1.1 mil m^3 (1991) to over 5.5 mil. m^3 (1995) of wood each year. The importance of this pest began to increase during the period 1992-1993, as a result of warm and dry weather and a huge number of trees blown down by wind, mainly in Austria, but in other central European countries as well. *Ips typographus* is usually considered to be a secondary pest, however after population densities increase, it becomes a primary pest. The main factors that predispose mature spruce stands to outbreaks are windthrow, snowbreaks, dry and hot weather during spring and summer, inadequate forest hygiene, and air pollution. Trees are attacked initially at the lower part of the crown. Trees which are located at the forest edge are stressed by sunlight mainly and are very susceptible to attack. Symptoms of attack include wood and bark dust from entrance holes produced by males, color change in needles from dark green to yellow, to orange, and then to brown, and loss of bark in the lower part of the crown.

Systematics and description: *I. typographus* belongs to the Family Scolytidae and Subfamily Ipinae (Tribe: Ipini). Its length is in the range of 4.2 - 5.5 mm, colour is dark brown and shiny. The most important characteristic is the presence of 4 teeth on the back part of each elytrum (total = 8 teeth). The elytrum is covered by deep holes in rows. The area between rows is shiny and without holes. Closely related species include *Ips amitinus* Eich., *Ips cembrae* He. and *Ips dupicatus* Sahl. All these species have 8 teeth on the elytra which is also covered by deep holes in rows, however the area between rows also contains holes.

The life cycle is different in lowlands (below 800 m) than in the highlands (over 800 m). The swarming of the first generation beetle begins in the lowlands at the end of April-beginning of May, and 2 weeks later in the highlands. The 2nd generation begins irregularly in August depending on the weather. A 3rd generation occurs irregularly in the lowlands only if the weather is warm and dry during spring and summer. Population densities of *I. typographus* can increase rapidly. If three generations occur in one year, populations can increase from one female and two males to 192,000 eggs under optimal conditions. Mature spruce trees can be killed by the occurrence of 2-3,000 beetles. In Europe, acceptable host trees include *Picea abies*, *Picea omorica* and *Pinus sylvestris*, whereas in Asia, *Picea obovata*, *Picea jezoensis*, *Pinus sibirica*, *Pinus koraensis*, and *Abies sibirica* serve as hosts.

Population density is assessed by using pheromone traps and trap trees. In the Czech and Slovak Republics three levels of population are determined by using the following criteria:

Method	Stable Population	Increasing Population	Outbreak Population
Pheromone Traps	fewer than 1,000 specimens/trap	between 1,000-4,000 specimens/trap	over 4,000 specimens/trap
Trap Trees	fewer than 0.5 entrance holes/1dm^2	between 0.5-1.0 entrance holes/1dm^2	over 1.0 entrance hole/1dm^2

The most endangered stands, that is those with the highest outbreak potential, are homogenous spruce monocultures. The least endangered stands are mixed broad-leaf-coniferous forests that contain beech (40-60%), silver fir (5-15%), spruce (20-40%) and other broad-leaf species (5-10%). The best approach for control of *I. typographus* is integrated pest management (combination of all known methods). These include forest hygiene, the use of mass trap trees, pheromone trapping, and the combination of trap trees and pheromones. A modification of the latter method, referred to as the "Swedish method," involves the placement of pheromone dispenser on trees that have been poisoned. The best prevention against the development of outbreak populations is to process rapidly windthrown and snowbreak trees, and trees that are initially attacked by *I. typographus*.

BEING A GOOD ECOLOGICAL SURGEON: USING BIOLOGICAL CONTROL SAFELY TO SUPPRESS DAMAGE FROM INVASIVE SPECIES

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ABSTRACT

Invasions of exotic species threaten economic and ecological resources of invaded regions and degrade biodiversity of invaded communities. Invasive species threaten native species by direct antagonism (herbivory, predation, pathogenicity), competition, or modification of invaded habitats. Examples of invasive species in the United States that illustrate these outcomes include the invasive herbivorous insects hemlock woolly adelgid (*Adelges tsugae* Annand), beech scale (*Cryptococcus fagisuga* Lindinger) which attack native trees, purple loosestrife (*Lythrum salicaria* [L.]) which invades native wetlands and competes with native plants for space in the habitat, zebra mussel (*Dreissena polymorpha* Phallas) which competes with native unionid mussels, and saltcedar (*Tamarix* spp.) in the southwestern deserts, which causes riparian water tables to drop, making areas unsuitable for most native plants. Responses to damage from invaders range from doing nothing (and accepting the damage, which may be increasing), habitat management (which may be applicable to some species and habitats, but not to others), to active control efforts. Active control efforts begin with prevention, followed by eradication efforts if new infestations are detected early, and then move to suppression. Techniques for suppression of established exotic species include mechanical, chemical and biological control. The first two approaches (mechanical and chemical) are especially useful when areas on which the invader is to be controlled are small, either because the total area infested is small, or because control is only desired in limited "specimen habitats," such as nature reserves of limited acreage. Problems associated with these methods (cost, mechanical disturbance, chemical pollution) increase significantly as the total infested area to be treated increases. In contrast, biological control is poorly suited to control exotic species that occur only on limited acreage and are not spreading because costs associated with biological control are large and occur at the beginning. However, biological control is well suited to control invasive species that occupy large areas because the cost of treatment does not increase in direct proportion to the size of the infested area. This cost savings occurs because released natural enemies reproduce and spread without direct inputs from managers. Safe use of biological control agents requires social consensus on several points. First, there must be broad agreement that the target species should be reduced in density and range over the whole of the region to which the biological control agents will spread. Second, adequate host range data on the natural enemies to be released must be available to estimate the spectrum of nontarget species likely to be affected by the biocontrol agents. Finally, a social judgment must be made that the estimated host range of the agents is acceptable, given the importance of the pest and the likelihood of effects on various specific nontarget species.

ANT COMMUNITIES AND ACTIVITIES IN THE MONONGAHELA AND GEORGE WASHINGTON NATIONAL FORESTS

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ABSTRACT

Ant communities and activities in Monongahela National Forest (MNF) (Pocahontas Co., WV) and George Washington National Forest (GWNF) (Augusta Co.,VA) were studied in 1995 and 1996. Nine 200 ha plots were established in each forest. Three parallel, 80 m long, 20 m apart ant sampling transects were marked out in each plot. Nine honey and peanut butter bait traps were placed on one of the transects each week on Mondays and collected on Tuesdays from early May to mid-August. Traps were put on alternative transects on each plot, so that each marked transect of a plot would receive baited traps every three weeks. Ants collected by the baits were identified and counted in the lab. A total of 29 species of ants were recorded, six of the species are rarely encountered and one was a new species. *Aphaenogaster rudis* were the most dominant ants in both forests (45.5%). Nine other species form a subdominant group (1%<each sp.<15%). Multivariate analysis showed that the species compositions between the two forests were significantly different. This correlates with the difference in soil moisture and vegetation type between the two forests. GWNF plots are more diverse in ant species composition. They had more *Camponotus pennsylvanicus* (De Geer), *Formica neogagates* Emery, *Tapinoma sessile* (Say), and *Myrmica* n. sp.1. Total number of species trapped per plot in the GWNF are higher than in the MNF plots. There is also a significant lag in early ant activity between the two forests. The ants in the MNF reached full activity in the middle of June, which is about 4-5 weeks later than in GWNF . This coincided with the later foliage development on the MNF. Several species showed a seasonal pattern in their activity. Among these, *Prenolepis imparis* (Say), *F. subsericea* Say, *Leptothorax longispinosus* Roger and *L. curvispinosus* Mayr were active in spring and early summer, whereas *Camponotus americanus* Mayr started to appear in early summer. Although 1995 and 1996 are greatly different in climate (dry and wet respectively), the ant fauna trapped by bait in 1995 and 1996 are very similar, which indicates the climate does not have immediate dramatic impact on ant composition.

EFFECTS OF WEATHER AND *ENTOMOPHAGA MAIMAIGA* RESTING SPORE ABUNDANCE

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ABSTRACT

The impact of *Entomophaga maimaiga* on gypsy moths (*Lymantria dispar* (L.)) is dependent on the presence of adequate moisture. However, because of differential susceptibility of gypsy moth larvae due to variability in their behavior at different ages, the timing of moisture availability in relation to developmental stage of the insect is probably more important than the actual amount of rainfall. A computer model that uses daily maximum-minimum temperatures and rainfall data from weather stations to calculate a number representing the susceptibility of the gypsy moth to *E. maimaiga* was used to look at spatial and temporal variations in this susceptibility. For the year 1989, when the fungus was first known to be established in North America, this susceptibility rating was determined for the New England States plus New York, Pennsylvania, and New Jersey. According to the model, weather patterns in Connecticut, southern New York, and northern Pennsylvania were most favorable for development of the fungus. The known distribution of *E. maimaiga* during that year was centered in Connecticut, suggesting that the infestation may have started in that state. When the yearly infection potential was calculated for weather stations in Connecticut from 1969 to 1996, it is clear that 1989 had the most favorable weather conditions for development of *E. maimaiga*. However, even though later years were not as favorable, it is nevertheless true that since 1989 *E. maimaiga* has been consistently abundant in gypsy moth populations. This is probably because many resting spores were deposited in the environment during 1989. It is now known that some resting spores may germinate at least 7 years later. Thus, these resting spores represent a huge, long lasting reservoir of fungus infection. Any attempts to understand the long-term impact of *E. maimaiga* on gypsy moth populations need to account for this large influence of resting spores as well as the yearly weather patterns.

GYPSY MOTH PARASITOIDS

IN THE DECLINING OUTBREAK IN LITHUANIA

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ABSTRACT

To determine natural enemies (parasitoids) most effectively regulating gypsy moth (*Lymantria dispar*) in Lithuania, gypsy moth larvae were collected from two declining populations during the development season in 1995-97 and reared individually on artificial diet in the laboratory. Collecting sites were chosen within naturally ceasing outbreaks in birch (*Betula pendula* Roth., *B. pubescens* Ehrh.), alder (*Alnus glutinosa* Gaerth, *A. incana* Dc.), and mixed birch-alder forests. Gypsy moth population density, assessed by routine egg mass (EM) count, was: 3791 ± 1328 EM/ha one year after peak outbreak (site 1, 1997); 829 ± 282 and 911 ± 382 EM/ha two years after peak (site 2, 1995 and site 1, late 1997, respectively); 45 ± 6 EM/ha three years after peak (site 2, 1996) and 3 ± 1 EM/ha four years after peak (site 2, 1997). Forty larvae were collected each week at site 1 (two plots, 4.8 ha total) in 1997 and 160 each week at site 2 (8 plots, 5 ha each) in 1995-96.

Insect parasitoids killed 25.0 to 36.5% of the 3,392 larvae that were reared (33.5 to 56.4% died from diseases; 8.5 to 30.2% developed to adult). Mean percent parasitism for each larval stage was: L1 = 3.1 ± 0.8%, L2 = 20.1 ± 1.5%, L3 = 39.4 ± 1.6%, L4 = 38.8 ± 2.0%, L5 = 52.7 ± 2.4%, L6 = 72.5 ± 2.9%.

Parasetigena silvestris R.D. (Diptera: Tachinidae) was the most abundant species (49.5% of all recorded parasitoids), causing 16.8% mortality, particularly in late instar larvae. *Phobocampe disparis* Vier. (Hymenoptera: Ichneumonidae) parasitized 7.4% of larvae, primarily in the 3rd (L2-L4) instar. *Meteorus pulchricornus* Wes. (Hymenoptera: Ichneumonidae) was responsible for 1.5% mortality of early instars (L2-L4). All other species of parasitoids caused 1% or less of gypsy moth mortality. *Siphona borealis* Mesner, a rare North European tachinid, was identified as a parasitoid of *L. dispar*; this is the first record of *L. dispar* serving as a host for this species.

USDA Interagency Gypsy Moth Research Forum
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