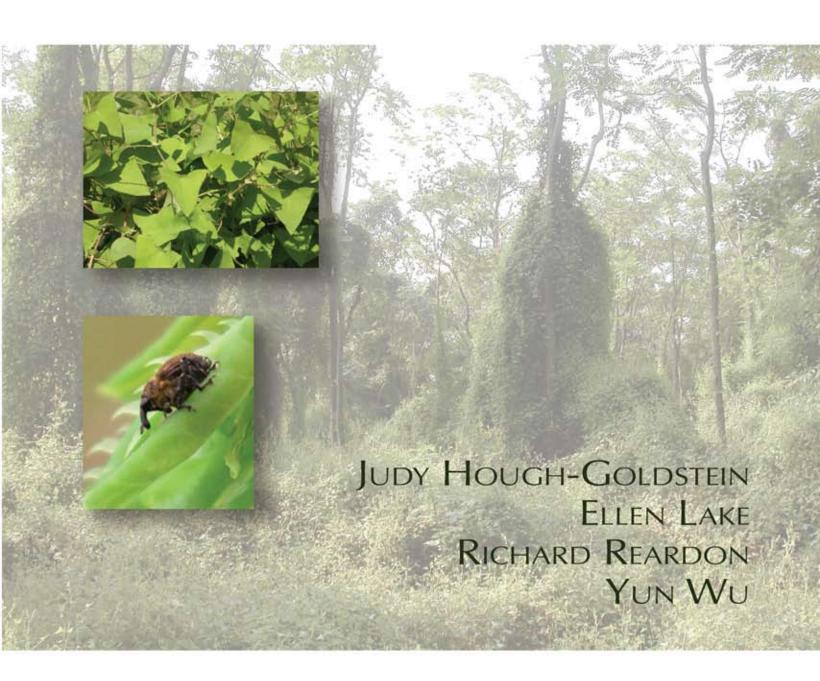
# BIOLOGY AND BIOLOGICAL CONTROL OF MILE-A-MINUTE WEED









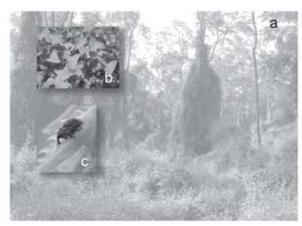


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Pesticides used improperly can be injurious to humans, animals, and plants.
Follow directions and read all precautions on the labels. Consult your local forest pathologist, county agricultural agent, or State extension agent about restrictions and registered users of particular pesticides.



#### Cover photo

a) Mile-a-minute weed infestation, Judy Hough-Goldstein; b) mile-a-minute weed, Judy Hough-Goldstein; c) orange weevil, Ellen Lake.

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# BIOLOGY AND BIOLOGICAL CONTROL OF MILE-A-MINUTE WEED

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#### Introduction

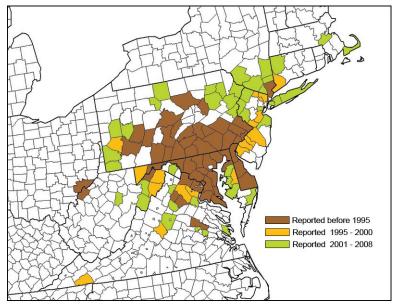
#### **Overview**

Mile-a-minute weed (MAM), Persicaria perfoliata (L.) H. Gross (Fig. 1), is a member of the family Polygonaceae. It is an annual vine that can grow up to 6 meters long over the course of a season. It is widely distributed throughout east Asia, including Japan, China, Korea, India, Indonesia, Bangladesh, Siberia, Philippines, Malay Peninsula, Indochina Peninsula, Nepal, and Turkey (Wu et al. 2002). It was introduced to the northeastern United States in the mid-1930s



Figure 1. Landscape infested with mile-a-minute weed.

from Japan, probably as seed unintentionally mixed in with holly seeds, and has since spread to ten states and the District of Columbia (Fig. 2).



**Figure 2**. Counties in the United States in which mile-a-minute occurs.

Mile-a-minute invades disturbed areas, such as roadsides, stream banks, rights-of-way, openings in forested areas, and regeneration areas, and crowds out most native vegetation. At high densities it can create monocultures. In addition to the loss of native biodiversity, with its stems and leaves covered with recurved spines, MAM is bothersome to people and their pets during outdoor activities.

The seed remains viable in the seed bank in the soil for six years, so managing MAM successfully depends on yearly treatments. Herbicides and hand-pulling plants can be effective management methods, but these are difficult to accomplish on a landscape with intermittent MAM populations.

The biological control program for MAM began in 1996. That year, the USDA Forest Service, Forest Health Technology Enterprise Team (FHTET), together with the University of Delaware and the Chinese Academy of Agricultural Sciences, initiated surveys for natural enemies and host-range studies in China and the U.S. In 2001, a colony of the weevil *Rhinoncomimus latipes* (initially misidentified as *Homorosoma chinensis*) was established in the USDA Agricultural Research Service (ARS) quarantine facility in Newark, Delaware, to study its biology and life cycle. Host-range studies were initiated with input from the Technical Advisory Group (TAG), which represents the interests of a diverse group of Federal and other agencies. A petition for release in the U.S. was submitted to the USDA Animal and Plant Health Inspection Service (APHIS) in 2003, and approved in 2004. The New Jersey Department of Agriculture began mass rearing the weevil in 2004 and the first release was made in Delaware that same year. Subsequent releases have been made in New Jersey, Maryland, Pennsylvania, and West Virginia. The search for additional natural enemies of MAM continues in China and Japan.

# **Biological control of weeds**

Problems caused by exotic invasive plants have increased dramatically in recent decades. In the US, it is estimated that invasive plant species comprise from eight to 47 percent of the total flora of most states (Rejmánek and Randall 1994). Many possess characteristics that favor their population increases, and have no natural enemies in their invaded range. So, once they become established, they are not easily suppressed or eliminated.

Classical biological control involves reconnecting exotic plants with the specialized natural enemies from their native ranges. This process begins with surveys in the target plant's area of origin to discover candidate natural enemies, progresses through studies of the candidate's biology and host specificity, and culminates with the release and evaluation of a candidate's damage to

the target plant. Damages will either limit weed reproduction or facilitate secondary infection by pathogens, which in turn will reduce the weed's ability to compete with other plants. In the eastern United States, projects have targeted aquatic, pasture, and forest weeds.

Biological control agents cannot be retrieved once they are released; therefore, they must be carefully selected and extensively studied before being approved for release (Wilson et al. 2004). The question arises as to what these specialized enemies will eat once they have reduced the target-weed population. Specialist insects have evolved over thousands of years to deal with specific secondary plant chemicals in their hosts, and cannot easily expand their range to feed on other plant species. Where host-range expansions have occurred, they generally have involved feeding on plant species that are closely related to the target weed. Therefore, it is critically important to determine, *prior to release*, whether any closely related desirable plant species occur in an area where a release is planned. If so, *prior to release*, it should be determined whether the insect can feed on those species. However, because even the most effective biological control agent will only reduce, not eradicate, the target-weed species, the long-term goal of any release is for both plant and insect populations to persist, but at relatively low levels.

There are advantages and disadvantages to classical biological control of weeds:

#### Advantages

- It is selective against a specific weed or closely related group of weeds.
- It can provide long-term control.
- Agents can disperse to areas not accessible to humans or equipment for control.
- The biological control agents are self-perpetuating, so there are no recurring acquisition, rearing, and reintroduction costs.

#### Disadvantages

- There are high initial program costs.
- It is not certainthat the agents will be effective (even effective agents will not work in every situation).
- There is a risk of unintended, adverse impacts on other plant species (non-target effects).
- Impacts may not be noticed for five to ten years.

The United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Quarantine (USDA-APHIS-PPQ) and the Canadian Food Inspection Agency (CFIA) are responsible for authorizing the importation of biological control agents into their respective countries. Federal laws in the U.S. and regulations are in place to minimize the risks to native plant and animal communities associated with introductions of exotic organisms to manage weeds. The Technical Advisory Group for Biological Control Agents of Weeds (TAG) is an expert committee with representatives from regulatory agencies, federal land management and environmental protection agencies from the United States, Canada and Mexico. TAG is concerned with the safety and potential impacts of prospective biological control agents. To that end it reviews all petitions to import new agents into the United States and makes recommendations to USDA-APHIS. Weed biological control researchers work closely with USDA-APHIS-PPQ and TAG to assess the environmental safety of potential weed biological control agents and programs. The Canadian counterpart to TAG is the Biological Control Review Committee (BCRC) (Bourchier et al. 2006).

In addition, each state in the United States has its own approval process to permit field release of weed biological control agents.

#### About this manual

This manual provides background information on mile-a-minute weed and the biological control insect *Rhinoncomimus latipes*, and provides guidelines for the use of biological control as either a stand-alone tactic or as a component in an integrated MAM management program. The chapters are:

Chapter 1 provides a detailed description of MAM, including taxonomy, description of the leaves, stems, flowers, seeds, and habitat, life history and occurrence in the United States.

**Chapter 2** provides the results of surveys for natural enemies of MAM in the US, Japan and China. It describes the weevil *R. latipes*, its biology and host range studies.

**Chapter 3** describes the mass-rearing, releases and spread of *R. latipes* and its impacts on MAM in the US.

Chapter 4 includes elements of a biological control program as well as an integrated management program for MAM.

Glossary defines technical terms essential in using and communicating about MAM biological control.

**References** provide critical literature on MAM biology, ecology, and biological control. Only publications cited directly in this manual are listed.

#### **Appendices**

- A. Mile-a-minute weed monitoring protocol and forms
- B. Mile-a-minute weed quick monitoring protocol and form

### CHAPTER 1: GETTING TO KNOW MILE-A-MINUTE WEED

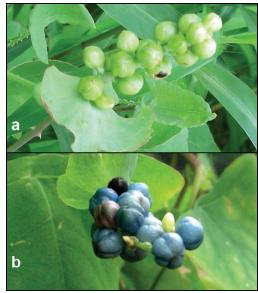
# **Description and classification**

Mile-a-minute weed is an herbaceous, annual vine with stems that grow up to 6 meters long in one growing season. It has triangular leaves, and its stems, petioles and leaf veins are covered with small, backward-projecting, recurved prickles. Leaves are alternate, simple, and 2.5 to 7.5 centimeters long and wide. Ocreae (fused stipules that surround the stem at each leaf node) are found in many species in the family Polygonaceae; in MAM they flare widely into a saucer

a

**Figure 3**. Mile-a-minute weed. Note triangular leaves (a), backward-projecting spines (b), and flared ocreae surrounding stems (c).

shape (Fig. 3). Flower buds, and later flowers and fruits, develop at the terminal tips. Flowers are small, green, and generally inconspicuous. The flowers give way to clusters of green berry-like fruits, which turn an iridescent bluepurple when mature (Fig. 4). Each fruit encloses a single, hard, shiny, black, seed, or achene.



**Figure 4**. Immature (a) and mature (b) berrylike fruit clusters.

Mile-a-minute weed was long classified in the large genus *Polygonum*, as *P. perfoliatum* L. However, more recently most botanists in North America have agreed that this plant should be placed in the genus *Persicaria* (Hinds and Freeman 2005). Along with other "tearthumbs" (all of which have recurved prickles on their stems), this species is in the section Echinocaulon, and the scientific name is now *Persicaria perfoliata* (L.) H. Gross. The North American species that are most closely related to *P. perfoliata* are *Persicaria sagittata* (L.) H. Gross (arrow-leaf tearthumb), and *Persicaria arifolia* (L.) Haraldson (halberd-leaf tearthumb). Also, the smartweeds, which include both native and introduced species, are now placed in the genus *Persicaria*.

# **Life history**

In the Northeastern United States, seeds germinate beginning in March or April (Fig. 5). Flowering begins in June or July and fruits may be produced beginning any time from June through August, probably depending on both site and weather conditions. Achenes are dispersed through human activities and by water, birds, deer and other mammals (see "Dispersal of mileaminute seeds by deer," below).



Figure 5. Mile-a-minute seedlings in early spring.

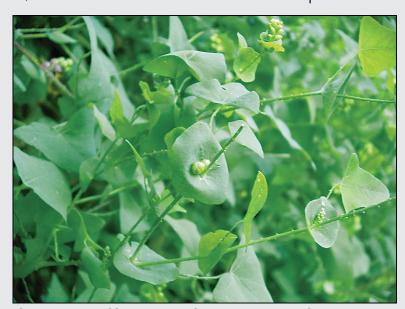
Ripe fruits not consumed by animals drop to the soil and many germinate under old plants the following spring. The seeds must go through a period of moist cold before they will germinate. Seeds can survive for more than one season and retain viability (see "Mile-a-minute seed bank persistence and viability," below). As an annual plant, the entire MAM plant dies with the first hard frost, generally in late October or early November in the Mid-Atlantic region.

# Dispersal of mile-a-minute seeds by deer

Mile-a-minute (MAM) seed dispersal and germination can be facilitated by white-tailed deer, *Odocoileus virginianus* Zimm. Deer can consume large numbers of a wide variety of seeds while they forage (Myers et al. 2004, Vellend 2002). They may travel substantial distances before defecating, thus transporting seeds hundreds to thousands of meters, and even farther during seasonal migration. In one study, 64% of the plant species that germinated from seeds present in deer pellet samples were from non-native species (Myers et al. 2004).

As MAM plants mature, seeds ripen and plant stems get woodier near the terminals. Plants in the field appear to hold the seed clusters up and out over the mat of vegetation (personal observation, E. Lake). Often, these terminal fruit clusters are missing (Fig. 6), with only an ocrea and part of the stem left behind. Erica Dale and Ann Herzig (Bryn Mawr College, unpublished data) collected deer scat and searched the samples for MAM

seed. Although large numbers of MAM seed fragments were found, many seeds also passed through the gut intact. In 18 deer pellet groups collected in the fall of 1997 and 1998, an average of 17.6 intact MAM seeds were found per pellet group (range 1–111). A germination experiment demonstrated that 40% of MAM seed scarified via passage through deer gut was viable.



**Figure 6**. Animal browse on mile-a-minute terminal.

# Mile-a-minute seed bank persistence and viability

Judith A. Okay, Riparian Specialist, Virginia Department of Forestry and Chesapeake Bay Program, Annapolis Maryland

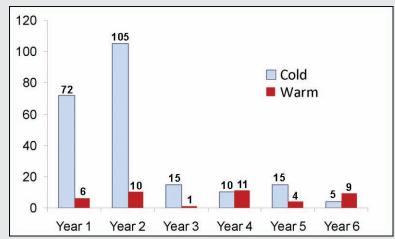
To assess *P. perfoliata* seed bank longevity and persistence, two experiments were conducted using achenes collected in the 1997 growing season. The first was a temperature-controlled experiment using refrigeration to induce germination, and the second involved achenes buried in soil. Each of the tests ran from September 1997 through July 2003.

#### **Temperature-controlled test**

A total of 264 achenes were placed on moist sponges in petri dishes in groups of about ten per dish. They were kept in an incubator without lights, and exposed to temperatures that simulated seasonal temperature changes, i.e. 35 to 37° F through fall and winter (October through April), and 65 to 68° F through spring and summer (May through September).

Sponges were kept moist, and achenes were checked weekly for germination, defined as the protrusion of the radicle through the seed coat.

The majority of the seeds germinated during the period when they were exposed to cold temperatures, and most germinated during years one and two (Fig. 7). However, a small number of seeds continued to germinate each year through year six. By the end of year six, more than 99% of the seeds had germinated.



**Figure 7**. Numbers of mile-a-minute seeds germinating when kept cold  $(35 - 37^{\circ} \text{ F, Oct.} - \text{April})$  and when kept warm  $(65 - 68^{\circ} \text{ F, May - Sept.})$  from a single batch of 264 mile-a-minute weed achenes collected in 1997 (Year 1 = Oct. 1997 – Sept. 1998).

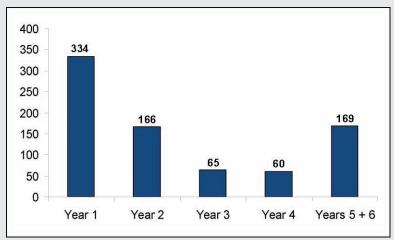
#### **Buried-seed test**

In October of 1997, 800 achenes were placed in four mesh bags (200 per bag), marked with orange survey flagging and buried side by side in a 3' x 3' plot at a depth of 5 to 6 inches in natural loamy-clay soil. The achenes were not watered or tended, but were left in the soil under natural conditions until the following spring. The mesh bags were exhumed each spring in late May or early June after a flush of *P. perfoliata* seedlings had emerged in the area of the test plot, indicating most germination had ceased. This was done each year from 1998 through 2001, and again in 2003. Undamaged achenes that had not germinated were separated from roots and opened husks of germinated achenes, counted, returned to the mesh bags, and reburied.

Continued, next page

# Mile-a-minute seed bank persistence and viability, continued

Of the 800 achenes buried in fall of 1997, over 40% germinated the following spring (Year 1, Fig. 8), and an additional 21% germinated the second year after burial. Most of the remaining seeds germinated at a lower rate over the next four years. By 2003 (Year 6), 99.3% of the buried seeds had germinated.



**Figure 8.** Number of seeds that germinated under natural conditions, buried in mesh bags and exhumed each year (not checked in year 5).

#### **Conclusions**

The seasonal dormancy observed in both the temperature-controlled test and the buried-seed test is a common response for summer annuals, which produce seeds that generally go dormant in response to the high temperatures of late summer and early fall, and germinate only during the cooler conditions of early spring. The need for a period of cold-wet stratification to break seed dormancy in *P. perfoliata* has been shown by others (summarized by Colpetzer and Hough-Goldstein, 2004), but this is the first test to show this pattern continuing over multiple years with a single batch of seeds exposed sequentially to a 7-month cold period and a 5-month warm period.

In both experiments, *P. perfoliata* seed persisted and remained viable in the seed bank for 6 years following collection, although most of the seed germinated during the first and second year. Van Clef and Stiles (2001) reported 32.6% viability of MAM seed that had been buried for three years, but did not test longer periods. The results presented here suggest natural resource managers attempting to control *P. perfoliata* should plan to continue control efforts for at least six growing seasons, because viable seed is likely to persist in the seed bank for at least that long.

#### **Distribution**

Mile-a-minute weed is indigenous to, and widely distributed in, Asia. It was first reported in the United States near Portland, Oregon, in the 1890s, but apparently did not persist west of the Rocky Mountains. The plant was introduced into the eastern U.S. in the mid-1930s at the Gable Nursery in Stewartstown, Pennsylvania, probably with holly seeds from Japan (Moul 1948). Analysis of random amplified polymorphic DNA (RAPD) profiles of MAM populations from China, Japan, Korea and the eastern U.S. support the suspected single introduction and Japanese origin of the eastern U.S. population (Shuppert 2001). No genetic variation was detected among populations in North America, suggesting an effectively clonal population. Specimens from the U.S. sites more closely resembled MAM from Japan than those collected in China and Korea, further supporting the likely Japanese provenance of the U.S. population.

Before 1980, MAM was limited to five counties in Pennsylvania and parts of Maryland. By 1995 it had been reported in 51 counties in seven states plus the District of Columbia (Fig. 2, above). An additional 19 counties, some in two new states, Connecticut and New Jersey, were added by 2000, and another 41 counties and one new state, Massachusetts, was added between 2001 and 2008 (Fig. 2, above). Other states in plant hardiness zones 6 and 7 are thought to be vulnerable to invasion by MAM in areas where adequate moisture is available (Okay 1997). It is not likely that the eastern U.S. population of MAM will progress into more tropical climates because those zones lack the cold vernalization period needed to break achene dormancy and stimulate germination.

In the U.S., MAM is a weed of parks, preserves, conservation easements, nursery crops, orchards, roadsides, drainage ditches and rights of way. Although it prefers low wet ground and full sun, it will tolerate semi-shade. Mile-a-minute appears to be more restricted to moist flood plains in Japan and China than in the U.S.

# CHAPTER 2: MILE-A-MINUTE WEED BIOLOGICAL CONTROL AGENTS

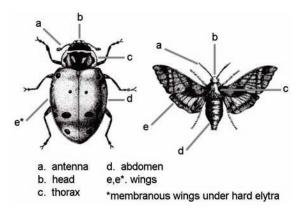
# **Basic insect biology**

Insects are a very large, diverse class of animals. Knowing basic insect anatomy and biology can help land managers recognize and identify biological control insects in the field. Adult insects have several unique characteristics: 1) an exoskeleton (external skeleton), 2) a segmented body comprised of three distinct regions: head, thorax, and abdomen, and 3) three pairs of legs (Fig. 9). Biological control agents for mile-aminute have a life cycle with four distinct stages - egg, larva, pupa, and adult (Fig. 10). This form of development is called complete metamorphosis.

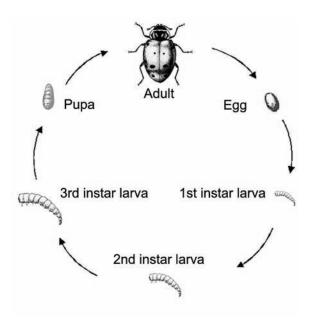
Immature insects also have an external skeleton that they must shed in order to grow. The process of shedding the exoskeleton is called molting. The stage of the insect between successive molts is called an instar. As larvae, insects generally complete three to five molts. The mature larva then molts into a pupa, the non-feeding stage when the insect changes from a larva to an adult.

# Insects found on mile-a-minute weed in the United States

One of the earliest surveys for natural enemies associated with mile-a-minute weed was conducted by Wheeler and Mengel (1984) in south central Pennsylvania in 1981 through 1983.



**Figure 9**. Generalized adult insect anatomy.



**Figure 10**. Complete metamorphosis.

They recovered more than 30 insect species (five orders, 15 families) that developed on MAM and 12 species that appeared to use the plant only for adult feeding. All 30 of these species

caused only minor herbivory. There were no leafminers, stem borers, internal fruit feeders, or gall makers. Three relatively oligophagous species were identified: *Lithacodia* [now *Pseudeustrotia*] carneola Guenee (Lepidoptera: Noctuidae), *Calothysanis amaturaria* Walker (Lepidoptera: Geometridae), and *Ametastegia* sp. (Hymenoptera: Tenthridinidae). All three species were rare.

Two surveys for native natural enemies of MAM were initiated in the late 1990s in the eastern U.S. in an effort to identify native natural enemies of MAM and their relative effectiveness (prior to the release of exotic species of natural enemies). The first was an effort by Jim Fredericks, M.S. student at the University of Delaware. Fredericks surveyed MAM populations in White Clay Creek State Park in New Castle County, Delaware; Elk Neck State Park in Cecil County, Maryland; Eastern Neck Island in Kent County, Maryland; and Pennypack Park in Philadelphia County, Pennsylvania in 1997 (Fredericks 2001). He collected insects associated with MAM once a week from June through October. He collected a total of 35 insect species, 21 of which were not previously reported to be associated with the plant. No internal stem or seed feeders were identified, supporting the observations of Wheeler and Mengel. Fredericks did not recover the three oligophagous species recovered by Wheeler and Mengel. Fredericks attempted to rear *C. amaturaria* on MAM, but the larvae failed to feed and died.

The second effort was a broader survey of various habitats that documented the accumulation of natural enemy species and their associated damage on MAM and evaluated their potential as biological control agents. This effort was conducted in Pennsylvania, Maryland, Delaware, and Virginia, from 1997 through 2000. The results of this broader survey are reported here.

#### Materials and methods

In 1997, 37 sites of various sizes and habitats containing MAM were located by the State Departments of Agriculture or Forestry in Delaware (2 sites), Maryland (16 sites), Pennsylvania (8 sites) and Virginia (11 sites). The center of each site was marked with a 6-foot metal stake, a photo was taken to represent the density of MAM, and GPS coordinates were recorded. Additional data for each site, including abundance of MAM, habitat type, and other plant species growing in association with *P. perfoliata*, were recorded. Each site was visited once every two weeks from June through September and insects were either hand-picked or aspirated from MAM plants, or collected by shaking plants over a white sheet. Most of the insects were collected on the leaves; other parts of the plants were also examined in an attempt to recover root borers, stem borers, and internal fruit feeders. Type and severity of damage and the plant parts affected were also recorded. Attempts were made to keep immature Lepidoptera alive and rear them to maturity; adult Lepidoptera were placed in a kill jar, and all other insect specimens were placed in 70% ethyl alcohol (ethanol). Field collectors provided initial taxonomic identification to family prior to submitting the completed forms and insect specimens to research associates in the Entomology Department at West Virginia University, who, in turn and when possible, provided the initial identification to genus and species. Identifications to genus and species were then confirmed by taxonomic specialists, including Drs. Linda Butler (Lepidoptera), John Strazanac (Orthoptera), Dave Smith (Symphyta), Shawn Clark (Coleoptera), and Charles Bartlett (Hemiptera, suborder Auchenorrhyncha). Portions of the sample areas were monitored again in 1998 (24 sites), 1999 (19 sites) and 2000 (13 sites).

#### **Results**

During the four-year study, over 2,000 specimens were recovered from *P. perfoliata*, representing seven orders and 110 families. However, many of these were known to be nonherbivores. Abundantly recovered phytophagous species were the oriental beetle, *Anomala orientalis* Waterhouse (Coleoptera: Scarabaeidae); Japanese beetle, *Popillia japonica* Newman (Coleoptera: Scarabaeidae); and meadow grasshoppers, *Conocephalus* spp. (Orthoptera: Tettigoniidae). Table 1 shows insects recovered from *P. perfoliata* that were likely to be phytophagous on MAM, including polyphagous insects that are known to feed on *P. perfoliata*, polyphagous insects that are known to feed on *Persicaria* or *Polygonum* species, and very polyphagous insects that might feed on species in these genera or related plants. Seventeen species of insects were common to this survey and the one conducted by Wheeler and Mengel (1984). Larvae of the fall webworm, *Hyphantria cunea* (Drury), were recovered in both surveys in the United States and this was the only species also recovered from MAM in China and in Japan (Miura 2008).

Numerous insect species were observed on, or collected from, MAM, although most were not actually observed either feeding on or causing damage to *P. perfoliata*. Those few insect species that were observed damaging MAM plants were polyphagous species that either might or are known to feed on Polygonaceae or related plants. Of these species, the most abundantly recovered phytophagous species was the Japanese beetle, *P. japonica*, followed by, in decreasing order of abundance:

- tarnished plant bug, *Lygus lineolaris* (Palisot de Beauvois) [Hemiptera, suborder Heteroptera: Miridae]
- potato aphid, *Macrosiphum euphorbiae* (Thomas) [Hemiptera, suborder Sternorrhyncha: Aphididae]
- locust leafminer, Odontota dorsalis (Thunberg) [Coleoptera: Hispinae]
- a small stink bug, *Mormidea lugens* (F.) [Hemiptera, suborder Heteroptera: Pentatomidae]

During August, *P. japonica* adults were especially abundant on all of the sites in each of the four states. In some areas, the adults defoliated 80 to 100% of the MAM plants in a localized area as well as individual plants. The defoliated plants recovered and continued to grow and produce quantities of viable seed.

The habitat of each site was recorded as being either partly sunny dry, partly sunny wet, full sun dry, or full sun wet. In its native range, *P. perfoliata* seems to persistently occupy wet sites (e.g., edges of creeks and rivers) whereas in this survey it occupied both wet sites and drier upland sites (e.g., roadsides, forest edges) (Table 1). In the upland sites, organic matter (leaves, plant material, etc) may be required to enhance seed germination and/or to keep the shallow root system moist and cool (Mountain, 1989). Many of the other plant species associated with MAM are also considered invasive weeds, including multiflora rose (*Rosa multiflora*), crownvetch (*Coronilla varia*), Canada thistle (*Cirsium arvense*), and garlic mustard (*Allaria petiolata*).

**Table 1.** Herbivorous insects collected from *P. perfoliata* in DE, MD, PA, and VA, 1997 – 2000.

Order (suborder)	0,, , , ; , ,	Relative	Part	sun	Full	sun	Other
Familya	Species	frequencyb	Wet	Dry	Wet	Dry	surveysc
Polyphagous species known to feed on mile-a-minute weed							
Coleoptera							
Chrysomelidae	Odontota dorsalis (Thunberg)	R	Х	Х			W
Scarabaeidae	Anomala orientalis Waterhouse	С			Х		
	Popillia japonica Newman	С	Х	Χ	Х	Х	W
Hemiptera (Heteroptera)							
Miridae	Halticus bractatus (Say)	R	Х	Χ			
	Lygus lineolaris (Palisot de Beauvois)	0	Х	Χ	Х	Х	W
Pentatomidae	Acrosternum hilare (Say)	R	Х	Χ	Х	Х	W
	Euschistus servus (Say)	R		Х	Х	Х	
	Euschistus tristigmus (Say)	R	Х	Χ		Х	W
Hemiptera (Auchenorrhyncha)							
Acanaloniidae	Acanalonia bivittata (Say)	R	Х	Х		Х	W
Cicadellidae	Graphocephala coccinea (Forster)	R	Х	Χ	Х	Х	W
	Graphocephala versuta (Say)	0		Х	Х	Х	W
Flatidae Metcalfa pruinosa (Say)		R	Х	Х	Х	Х	W
Hemiptera (Sternorrhyncha)							
Aphididae	phididae Macrosiphum euphorbiae (Thomas)		Х	Χ	Х	Х	W
Lepidoptera							
Arctiidae	Estigmene acrea (Drury)	R		Χ			W
	Spilosoma virginica (F.)	R		Χ		Х	W
Geometridae	Calothysanis amaturaria (Walker)	R			Х		W
Noctuidae	Palthis asopialis (Guenée)	0	Х	Х		Х	
Orthoptera							
Acrididae	Melanoplus differentialis (Thomas)	R	Х	Х		Х	W
Tettigoniidae	Amblycorypha oblongifolia (DeGeer)	R	Х	Х		Х	W
	Amblycorypha rotundifolia (Scudder)	R					
	Atlanticus sp.	R	Х	Х		Х	
	Conocephalus brevipennis (Scudder)	R					
	Conocephalus sp.	С	Х	Х	Х	Х	
	Scudderia furcata Brunner	R	Х	Х		Х	W

<sup>&</sup>lt;sup>a</sup>Orders, suborders (Hemiptera only), and families as in Triplehorn and Johnson (2005).

<sup>&</sup>lt;sup>b</sup>Relative frequency: R, rare, taken at one or two sites in one state, usually in small numbers; O, occasionally collected at two or more sites in one or two states; C, common, taken at most sites in more than two states.

<sup>&</sup>lt;sup>c</sup>Other surveys: D: also listed as associated with mile-a-minute in China (Ding et al. 2004); W: also listed as associated with MAM in Pennsylvania (Wheeler and Mengel 1984).

Table 1, continued. Herbivorous insects collected from P. perfoliata in DE, MD, PA, and VA, 1997 – 2000.

Order (suborder) Family <sup>a</sup>	\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\			Full sun Wet Dry		Other surveys <sup>c</sup>	
Polyphagous species kno	wn to feed on Polygonaceae						
Coleoptera							
Chrysomelidae							
	Diabrotica undecimpunctata Mannerheim	R	Х	Х			W
	Diachus auratus (F.)	R	Х	Х		Х	
	Disonycha glabrata (F.)	R			Х		
	Luperaltica senilis (Say)	R	Х	Х	Х	Х	
Hemiptera (Heteroptera)			•		•	'	
Miridae	Halticus sp.	R	Х	Х		Х	
Thyreocoridae	Corimelaena sp.	R	Х	Х			
Lepidoptera						'	
Arctiidae	Pyrrharctia isabella (Smith)	R	Х				
Geometridae	Prochoerodes tranversata (Drury)	R		Х			
Tortricidae	Sparganothis sulfureana (Clemens)	R				Х	
Orthoptera			•			,	
Acrididae	Melanoplus sanguinipes (F.)	R	Х	Х		Х	
Tettigoniidae	Microcentrum sp.	R		Х			
Very polyphagous species	s that might feed on Polygonace	ae					
Coleoptera							
Chrysomelidae	Epitrix fuscula Crotch	R				Х	
	Oulema sayi (Crotch)	R	Х	Х		Х	
Curculionidae	Myllocerus hilleri Faust	R	Х	Х		Х	
	Otiorhynchus ovatus (L.)	R	Х	Х	Х	Х	
Hemiptera (Heteroptera)		,					
Berytidae	Jalysus sp.	R	Х	Х		Х	
	Neides muticus (Say)	R	Х	Х		Х	
Coreidae	Leptoglossus sp.	R	Х		Х	Х	
Cydnidae	Sehirus cinctus (Palisot de Beauvois )	R	Х	Х			
Miridae	Adelphocoris sp.	R	Х		Х	Х	
	Stenodema trispinosa Reuter	R	Х		Х	Х	
	Stenodema vicinum (Provancher)	R	Х		Х	Х	
Pentatomidae	Holcostethus limbolarius (Stal)	R				Х	
	Menecles sp.	R	Х	Х		Х	
	Mormidea lugens (F.)	R	Х	Х	Х	Х	
	Nezara sp.	R	Х	Х	Х	Х	
Rhopalidae	Arhyssus sp.	R			Х		

<sup>&</sup>lt;sup>a</sup>Orders, suborders (Hemiptera only), and families as in Triplehorn and Johnson (2005).

<sup>&</sup>lt;sup>b</sup>Relative frequency: R, rare, taken at one or two sites in one state, usually in small numbers; O, occasionally collected at two or more sites in one or two states; C, common, taken at most sites in more than two states.

<sup>&</sup>lt;sup>c</sup>Other surveys: D: also listed as associated with mile-a-minute in China (Ding et al. 2004); W: also listed as associated with MAM in Pennsylvania (Wheeler and Mengel 1984).

Table 1, continued. Herbivorous insects collected from P. perfoliata in DE, MD, PA, and VA, 1997 – 2000.

Order (suborder) Family <sup>a</sup>	Species	Relative frequency <sup>b</sup>	_	sun Dry			Other surveys <sup>c</sup>
Very polyphagous species	that might feed on Polygonace	ae, continued					
Hemiptera (Auchenorrhyncha)							
Cercopidae	Philaenus spumarius (L.)	R	Х	Х	Х	Х	
Cicadellidae	Draeculacephala mollipes (Say)	0	Х	Х	Х	Х	
	Oncometopia orbona (F.)	0		Х			
	Paraulacizes irrorata (F.)	R	Х	Х		Х	
	Tylozygus bifidus (Say)	R	Х	Х		Х	
Membracidae	Entylia carinata (Forster)	С				Х	
Hemiptera (Sternorrhyncha)							
Aphididae	Aulacorthum solani (Kaltenbach)			Х			
Lepidoptera							
Arctidae	Hyphantria cunea (Drury)	R		Х			D.W

<sup>&</sup>lt;sup>a</sup>Orders, suborders (Hemiptera only), and families as in Triplehorn and Johnson (2005).

#### **Discussion**

In this survey there was no evidence of seed or root feeders even though adults of several families of Coleoptera (e.g., Elateridae, Scarabaeidae) were recovered, and their immatures are associated with polyphagous root feeding. Many taxa were recovered from MAM foliage but few were associated with herbivory. Aphids were recovered on leaves and stems of many plants but the damage was minimal (less than 1%) on individual plants. Obviously, there has been an accumulation of taxa on MAM but at least 90% are transient or highly polyphagous.

*P. perfoliata* appeared to be equally abundant in moist and dry sites in this survey, although the quantity of organic matter might be a critical factor on the drier sites. In its native range, MAM is associated with moist sites; therefore, populations are probably regulated by seasonal flooding as well as natural enemies.

Under optimal conditions plants can compensate for the negative effects of herbivory; therefore, both the timing and duration of defoliation are important factors in regulating the host. *Popillia japonica* was the most abundant defoliator of MAM but had minimal impact on the survival and seed production of individual plants.

<sup>&</sup>lt;sup>b</sup>Relative frequency: R, rare, taken at one or two sites in one state, usually in small numbers; O, occasionally collected at two or more sites in one or two states; C, common, taken at most sites in more than two states.

<sup>&</sup>lt;sup>c</sup>Other surveys: D: also listed as associated with mile-a-minute in China (Ding et al. 2004); W: also listed as associated with MAM in Pennsylvania (Wheeler and Mengel 1984).

#### Insects found on mile-a-minute weed in Asia

#### China

In 1996, a collaborative project was initiated between USDA Forest Service, Forest Health Technology Enterprise Team (FHTET) and the Chinese Academy of Agricultural Sciences Institute of Biological Control (now Institute of Environment and Sustainable Development in Agriculture) to survey and screen biological control agents of MAM in China for possible release in the eastern U.S. Surveys for phytophagous insects were conducted from 1996 to 2001 in 23 provinces including some in northeastern China, where the climate is similar to that of the eastern United States, and southwest China, which is considered the center of origin of the family Polygonaceae (Ding et al. 2004).

A total of 111 phytophagous species from six orders and 29 families were associated with MAM in China. Although most were leaf feeders, several stem borers, fruit feeders, and seed feeders were found. No insects were recovered from the roots. Eleven of the species were regarded as important because either they cause severe damage on MAM or have a narrow host range (Ding et al. 2004). Included among the species collected were:

- the weevil Rhinoncomimus latipes Korotyaev (Curculionidae)
- three oligophagous leaf beetles, *Smaragdina nigrifrons* (Hope) (Eumolpidae), *Gallerucida bifasciata* Motschulsky and *Galerucella placida* Baly (both Chrysomelidae)
- a moth, Timandra griseata Peterson (Geometridae)
- a hemipteran, *Cletus schmidti* Kiritschenko (Coreidae)
- the sawfly, *Allantus nigrocaeruleus* (Smith) (Tenthredinidae)

## Japan

In 2004 and 2005, Dr. Kenji Fujisaki at Kyoto University initiated a survey for herbivorous insect fauna of MAM. Parts of Japan are in the native range of MAM (Ohwi 1965) and many of the survey sites are a good climatic match to the northeastern United States (Miura et al. 2008). Fujisaki conducted surveys at 15 sites from Kagoshima in the south to Sapporo in the north. They consisted of timed visual surveys (15 min per sample, two to six samples per site on a given sample date) with only one or two visits per year to most of the sites. A total of 50 herbivorous insect species were recovered on MAM:

- 26 Hemiptera (52%)
- 11 Lepidoptera (22%)
- 9 Coleoptera (18%)
- 3 Orthoptera (6%)
- 1 Hymenoptera (2%)

Six species appeared to be potential Polygonaceae specialists:

• 2 Hemipterans, the bug *Coptosoma parvipictum* Montandon (Pataspidae) and aphid *Trichosiphonaphis ishimikawae* (Shinji) (Aphididae)

- 2 Lepidopterans, *Timandra apicirosea* (Prout) (Geometridae) and *Oligonyx vulnerata* (Butler) (Noctuidae)
- 1 Sawfly, *Allantus luctifer* Smith (Tenthredinidae)
- 1 Beetle, *Rhinoncomimus latipes* Korotyaev (Curculionidae) (Miura et al. 2008)

Of the six specialist herbivores, *R. latipes* appeared to be the most promising natural enemy. This observation supports results from surveys conducted in China, as well as host-range testing, and the release of *R. latipes*, in the U.S. (Miura et al. 2008).

In 2006 and 2007, additional surveys for natural enemies of MAM were conducted by Dr. Naoto Kamata at the University of Tokyo. Twelve habitats with sites established along rivers or streams in the suburbs of the Tokyo Metropolitan area were monitored. Mile-a-minute weed appeared above ground in mid-May, began to decline in early October, and disappeared by mid-November. These sites were scouted for insects once or twice a week from the middle of May to the end of November. During each scouting session, at least 400 stems of MAM were inspected for 2 to 3 hours. In total, eight species of herbivorous insects were recovered on MAM in 2006: Allantus luctifer (sawfly), Hyphantria cunea (moth), Timandra apicirosea (moth), Cifuna locuples confusa (moth), Orgyia thyellina (moth), Helicoverpa armigera (moth), Apoderus erythrogaster (Attelabidae), and Rhinoncomimus latipes (weevil). Another moth, Spodoptera litura, was recovered in 2007. The weevil R. latipes was the most common herbivorous insect. It fed only on MAM in choice and no-choice tests. As was reported in the Fujisaki surveys in 2004 and 2005, the moth *Timandra apicisocea* was recovered in fairly abundant numbers in 2006 and 2007. It was dismissed as a potential biological control agent for release in the U.S. because, based on the results of no-choice tests, it also fed on common buckwheat (Fagopyrum esculentum Moench), tartary buckwheat (F. tartaricum Gaertn.) and rhubarb (Rheum rhabarbarum).

# Insects tested in the United States and China for host specificity

## Timandra griseata Peterson (Lepidoptera: Geometridae)

In August 1999, Ding Jianqing, with the Institute of Environment and Sustainable Development in Agriculture (formerly the Chinese Academy of Agricultural Sciences Institute of Biological Control) in Beijing, collected larvae and pupae of *Timandra griseata* from the field in Henan and Hubei provinces and sent them to the USDA-ARS Beneficial Insects Introduction Research (BIIR) quarantine facility in Newark, Delaware. *T. griseata* defoliated potted MAM, developing from egg to adult in approximately 26 days. However, its host range was considered to be too broad for it to be released in the United States, because it also fed and developed on common buckwheat (*Fagopyrum esculentum* Moench) and tartary buckwheat (*F. tartaricum* Gaertn), and accepted these species and MAM equally in choice tests (Price et al. 2003).

# Rhinoncomimus latipes Korotyaev (Coleoptera: Curculionidae)

Adults of *R. latipes* (initially misidentified as *Homorosoma chinensis* Wagner) were collected in Changsha, Henan province in China by Ding Jianqing and sent to the BIIR quarantine laboratory in July of 1999 and 2000 (Fig. 11). These weevils were found to have a relatively



Figure 11. Adult Rhinoncomimus latipes.

high reproductive rate and short generation time on potted MAM. Adults lay eggs on MAM leaves or stems. Eggs hatch in about 3 days (Price et al. 2003). Larvae quickly bore into stems at nodes, and feed internally (Fig. 12). Once fully grown, they crawl or drop to the soil where they pupate. The new adults emerge from the soil, crawl up nearby MAM plants, mate, and begin laying eggs.

In tests in China, R. latipes did not feed on 28 species of plants in 18 families outside of the Polygonaceae. Within the Polygonaceae, adult *R. latipes* did not feed on any plant other than MAM in choice tests or lay eggs on any species other than MAM, and larvae survived only on MAM. In quarantine in Newark, Delaware, R. latipes did not oviposit or complete larval development on two crop plants, buckwheat and rhubarb, within the family Polygonaceae, (Price et al. 2003). Subsequent tests were conducted on representatives from all of the Sections within the genus *Polygonum* sensu lato and on representatives of genera other



Figure 12. Larva feeding in stem.

than *Polygonum* within the family Polygonaceae, especially genera that contain threatened and endangered species. Also included were representatives of families thought to have chemical affinities with the Polygonaceae. Adult weevils in these tests fed and survived on a few species, but did not lay any eggs on plants other than MAM. In choice tests adults almost exclusively ate MAM, and newly hatched larvae placed on other plant species did not survive (Colpetzer et al. 2004). Based on these results, a release permit was granted by USDA-APHIS in July of 2004.

In the field in China, adults of *R. latipes* were collected from the upper petiole and the upper surface of the lamina, particularly on the first or second youngest leaves of MAM plants (Ding Jianqing, personal communication). Adults fed externally by scraping the epidermal layer and underlying cells, usually penetrating through to the other side of the leaf to form

# Host specificity testing

Matthew J. Frye, PhD student, University of Delaware

Host specificity testing of potential weed biological control agents is an essential step in determining the safety and efficacy of the insect or pathogen under evaluation. The primary objective of these tests is to determine the physiological host range of the agent, i.e. in addition to the target weed, which plant species from the introduced range are suitable for insect feeding, development, and reproduction.

The first step in host specificity testing is to develop a list of plants that may be at risk of damage from an imported phytophagous insect. This list must be reviewed by the Technical Advisory Group (TAG), an independent committee that reports to the USDA Animal and Plant Health Inspection Service. Test-plant species are selected based on their phylogenetic (evolutionary) relationship to the target weed, focusing primarily on species closely related to the target. The list of test plants also may include host plant species compiled from historical accounts of a potential agent, host plants of insects closely related to a potential agent, plant species that share morphological and biochemical traits or habitat requirements with the target weed, and crop and ornamental plants of economic value.

After a potential biological control agent has been selected from field surveys and preliminary tests in the native range of the target weed, the insect should be sent to a quarantine facility in the country where it is to be introduced for further evaluation. Included in the evaluation are no-choice tests in which insects are presented with a single, non-target, test-plant species at a time. Feeding, development, and survival rates are recorded and compared to those for insects on the target weed. No-choice oviposition tests are conducted to assess whether a female will oviposit (lay eggs) when confined to a single test plant. Tests used to determine the insect's host specificity may include choice tests, in which insects are presented with a combination of test-plant species along with the target weed, and their oviposition or feeding is recorded. Choice tests may include all plant species used by adults for oviposition as well as plant species from the no-choice tests fed upon by insects in any life stage.

a characteristic feeding hole. Newly hatched larvae bored into the young stem or bud from the top, and then tunneled downwards inside the stem. The combination of heavy defoliation by adult weevils and larval stem boring caused leaves to desiccate and curl until young shoots gradually withered away.

There are at least two generations of weevils per year in China. They overwinter as adults and emerge in early to mid-May when MAM vines are 12 to 15 inches long (Ding Jianqing, personal communication). High adult weevil populations have been observed in July, when they can be collected easily from MAM, often as mating pairs. Typically, three or four weevils per plant are found at this time, but in an exceptional year there could be as many as six to ten weevils per plant.

In culture in China, females began to oviposit 2 to 8 days after copulation, and continued to oviposit for 80 to 100 days. Tests with 25 pairs of adults showed that mean egg production was about 180 per female (Ding, unpublished data). No parasites were found in weevils collected as adults or in laboratory cultures. No insect pathogens were observed in the field or laboratory.

In quarantine in Newark, Delaware, the total development time (egg to adult) averaged 26 days, and egg production averaged about 130 eggs per female (Price et al. 2003). Adults from the previous year were observed to live through August and into late September in the laboratory, indicating that they can live up to 1 year. Adult *R. latipes* are black upon emergence, but turn orange-brown soon after feeding on MAM (Fig. 13).

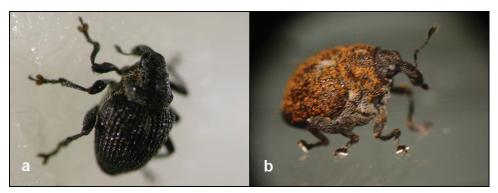


Figure 13. Black (a) and orange (b) weevils.

#### CHAPTER 3. RHINONCOMIMUS LATIPES IN THE UNITED STATES

# Mass rearing

Dan Palmer, Amy Diercks, and Caryl Ott

In August, 2004, mass rearing of *R. latipes* was initiated at the New Jersey Department of Agriculture's Phillip Alampi Beneficial Insect Laboratory (PABIL), in West Trenton, New Jersey. The lab was specifically designed for mass rearing insects. Rearing is on-going, with continual improvement in methods and efficiency.

It is well known that the quality of host material is paramount to successful insect rearing. Seeds collected from the field were initially used to propagate the plant. The seed-scarification process required for germination proved to be very time consuming, so vegetative reproduction was tried. It proved to be successful, and the use of cuttings from "mother plants" replaced the seed method of propagation. Standard growing procedures in the greenhouse were investigated to determine the needs of the plant. Good conditions include:

- fertilizer (Scotts<sup>TM</sup> General Purpose/Peter's Professional® ammonium nitrate fertilizer, 20-20-20), strength of 200 ppm
- day and night greenhouse temperatures of 75° F and 65° F respectively
- care in frequency of watering
- Fafard® mix for soil medium
- 6-inch plastic pots
- a 7-week-old plant

A healthy mother plant provides thick stems to use for cuttings. Replacing these plants approximately every 5 weeks ensures the supply of good cutting material and avoids greenhouse pests. The two lower nodes of the cutting (trimmed 0.25-inch below the lowest node and with the leaf cut off of both lower nodes) are soaked in rooting hormone for 10 seconds and then placed in plastic containers with individual compartments filled with a moist mixture of vermiculite and milled sphagnum moss (Fig. 14). The cuttings are placed in a shal-



**Figure 14**. Mile-a-minute weed (*Persicaria perfoliata*) cuttings.

low bin under fluorescent lights at 82° F and 100% humidity for 2 weeks. After a 3-day graduated venting process, the cuttings are transplanted into pots, watered, misted, and covered loose-

ly with plastic for 1 day. The plants are placed in the greenhouse four days after transplant. Watering is carefully monitored to avoid over-watering.

A precision pruning technique was developed to keep the plants at a manageable size while still providing the terminals (growing tips) needed for weevil reproduction. Female *R. latipes* lay most of their eggs on plant terminals; newly hatched larvae only burrow into the very young leaf nodes on a terminal. A plant with a sturdy base and five to eight thick-stemmed terminals is best-suited for insect production (Fig. 15).

Plants are kept under grow lights through all insect rearing stages to optimize plant quality leading to increased



**Figure 15**. "Ideal" pruned mile-a-minute plant for weevil rearing.

weevil production. Room temperatures are kept at 80° F with 16L: 8D lighting conditions. The Tek-5 grow lights are used to maintain the temperature inside the containers at about 82° F. The humidity inside both the egg laying and the development containers is between 95 and 100%. For egg laying, mating pairs of *R. latipes* are placed on seven-week-old MAM plants inside the containers (Fig. 16). Eggs start hatching on day three or four. Every 2 to 3 days, the plants are moved to development containers (large plastic bins) and new plants are added to the egg-laying containers (Fig. 17).



Figure 16. Egg-laying container.



Figure 17. Development containers.

As the eggs hatch, the larvae burrow into the youngest leaf nodes and develop through all larval instars within the nodes and stems. Because many of the leaf nodes occupied by the larvae are new growth that began at about the time the eggs were laid, grow lights are needed over the development bins. Seven or 8 days after egg hatch, the mature larvae chew their way out of

the nodes and either crawl or drop to the moist soil medium below, where they pupate (Fig. 18). At this point the foliage inside the bin is replaced with a "trap plant." The pre-pupa builds a capsule around itself with the soil medium attached to the outside and pupates inside the capsule. After spending the pre-pupal and pupal stages in the soil, the adults emerge between day 17 and 20 and crawl up the trap plant to feed, where they can be collected using an aspirator.

The adults are either stored in a cage with an abundance of MAM plants or, if to be shipped soon, placed in a release cup with a honey-water sponge and kept at 55° F until shipped. Three methods are being evaluated for storing adults during the winter:



Figure 18. Rhinoncomimus latipes pre-pupa.

- Some adults are put in cages with large MAM plants and left outdoors through fall and winter to have the weevil go through a natural winter season.
- Some are put in a cage indoors with abundance of MAM plants, kept at 55° F, and brought out to feed at 74° F three times a week.
- Others are stored in moist sphagnum moss in Ziploc® bags at 34° F after pre-conditioning on MAM plant material, first for one week at 65° F, then for another week at 55° F, until feeding is reduced.

An organization could use similar procedures to rear *R. latipes* in its own facilities. The insects should be reared in a room kept at 80° F (or 82° F inside the container). The egg-laying containers can be clear plastic display boxes with a "no-see-um" netting covering a 1.5-inch hole in the top. The development containers can be polycarbonate clear plastic bins with three, 2-inch holes on each side covered with netting, and lids with three 2-inch holes covered with netting. The bins will keep the environment humid, so humidity inside the room may not be a concern. Grow lights over both oviposition and development containers are very important. The maximum rearing temperature inside the containers is around 88° F. If temperatures inside the containers drop below 78° F, the insects will have a longer life cycle.

# Release of Rhinoncomimus latipes in New Jersey

Mark Mayer

#### **Monitored sites**

In addition to mass rearing *R. latipes*, PABIL personnel have released the weevils at numerous sites in New Jersey, several of which have been monitored using the "Mile-a-Minute Monitoring Protocol" developed by Dr. Judy Hough-Goldstein (Appendix A). Four field sites were set up for monitoring, three in Southern New Jersey (two at Floodgate Road in Greenwich, Gloucester County, and one at Department of Defense [DOD] Ponds Wildlife Management Area [WMA] in Pilesgrove, Salem County), and one in Central New Jersey at Pinelands Water and Wastewater Company in Vincentown, Burlington County. Weevils were released on two sites, and two were monitored as control sites. The control sites did not receive weevils, but MAM populations were monitored for comparison with release sites.

In spring, 2005, PABIL field personnel established two new sites in Salem County and dropped the Vincentown and the DOD Ponds sites, because these sites had been disturbed frequently by the public and there was a possibility that chemical control measures had been implemented. The new 2005 release site was at the Abbotts Meadow Wildlife Management Area in Elsinboro Township; the control site was located in the Supawna Meadows National Wildlife Refuge in Pennsville Township. One of the two Floodgate Road sites was retained as a release site and the other as a control site.

For all releases, weevils were brought to the field in 16-oz. wax-lined, hot-beverage Sweetheart® cups with holes cut into each end (Fig. 19). Nylon mesh was secured over the holes



Figure 19. Field release of Rhinoncomimus latipes.

and a Pioneer plastics® Petri dish containing a sponge moistened with honey and water was taped to the bottom of the cup. Excelsior was placed in the cup to give the weevils more resting sites. Upon release, the excelsior and any weevils on it were removed from the cup and placed gently on the MAM. The cup was placed in the MAM to allow the rest of the weevils to walk out on their own.

Weevil counts can be misleading, because they tend to drop, undetected, from the plant when disturbed during the survey process. Often, weevils can be found by first looking for feeding damage near the release site (Fig. 20), and then by searching for them on nearby leaves and terminals. Another sign of weevil activity in the field is the presence of damaged nodes (Fig. 21), indicating areas where larvae have bored into or out of stems. Although they are very tiny, the presence of weevil eggs (Fig. 22), with their characteristic peanut shape and thin covering of frass strips (insect fecal material), is another definitive sign of weevil activity. Foliage damage alone is not always adequate proof of weevil presence, because other organisms can also feed on MAM, notably Japanese beetles, which can be found on the plants in July and August (Fig. 23). Although not definitive proof of infestation, where weevils occur, feeding holes on MAM often make the plant stand out among other



Figure 20. Adult weevil feeding damage in early Figure 21. Damaged nodes, indicating larval feeding. spring.







**Figure 23**. Japanese beetles feeding on mile-a-minute.

plant species (including other closely related plant species), especially in early spring (Fig. 24).

The Floodgate Road release site in Gloucester County received 200 weevils in July 2004 and 3,297 in 2005. The weevils established quickly; the population grew so rapidly, the MAM was completely defoliated by October 2006 (Figs. 25, 26). There were so many weevils present at that site that more than 200 were collected in under a minute simply by putting a clipboard under the defoliated stems and gently tapping the plants (Fig. 27). The large numbers of weevils present in October 2006 indicated there was potential to establish field insectaries. To that end, in September of 2007, 200 weevils were collected from defoliated plants and redistributed on a site in Hunterdon County.



**Figure 24**. Early spring damage to mile-a-minute weed (note that the closely related *Persicaria sagittata* is untouched).



**Figure 25**. Mile-a-minute at Floodgate Road July 2004 (left), in October 2006 (middle) and October 2007 (right) after *R. latipes* feeding. Note the *Prunus* sp. bush in the foreground (middle) was not visible prior to weevil release in 2004 (left), because it was covered by mile-a-minute. The *Prunus* sp. grew once mile-a-minute was reduced.



Figure 26. Defoliation at Floodgate Road, October 2006.



Figure 27. Weevils on clipboard, October 2006.

Between 2005 and 2006, MAM seedlings were counted each spring in each of the ten permanent quadrats (see Monitoring Protocol, Appendix A) at the release and control sites (Table 2). Although the spring seedling count increased at Floodgate Road between 2006 and 2007, the MAM plants were significantly suppressed by the weevils in 2007. Over the course of the season, many MAM plants could not compete against other plant species, and seedling numbers were much lower in spring of 2008 (Table 2). The control site at Floodgate Road was so overrun with weevils migrating from the release site, it could no longer serve as a control site after 2006.

Following release of nearly 7,000 weevils between April and September, 2005, both the spring seedling counts and percent of MAM cover at Abbott's Meadow were dramatically reduced (Tables 2 and 3). In contrast, both seedling counts and percent cover remained high at the control site.

**Table 2**. Average number of mile-a-minute seedlings (± SEM) at release and control sites in New Jersey.

Site	Number of R. latipes released in 2005	Spring 2005	Spring 2006	Spring 2007	Spring 2008
Floodgate Road					
Release	3,297	290.4 ± 39.3	132.0 ± 25.5	207.6 ± 63.6	31.5 ± 10.4
Control		349.3 ± 52.0	161.6 ± 22.5		
Abbott's Meadow					
Release	6,976	401.0 ± 20.6	30.0 ± 12.4	93.5 ± 29.5	8.1 ± 5.9
Control		484.0 ± 73.6	505.2 ± 128.3	457.4 ± 87.7	192.8 ± 71.8

**Table 3**. Average percent cover of mile-a-minute (± SEM) at release and control sites in New Jersey.

Site	Number of <i>R. latipes</i> released in 2005	Spring 2005	Spring 2006	Spring 2007	Spring 2008
Floodgate Road					
Release	3,297	55.5 ± 5.9	47.0 ± 10.6	35.4 ± 10.3	12.1 ± 3.6
Control		60.0 ± 6.5	55.0 ± 7.7		
Abbott's Meadow					
Release	6,976	45.0 ± 3.1	4.2 ± 2.0	9.4 ± 4.9	1.8 ± 1.0
Control		60.5 ± 6.8	66.9 ± 7.7	77.5 ± 8.1	$37.6 \pm 8.7$

In 2006, a large number of *R. latipes* adults emerged after overwintering at Abbott's Meadow. Heavy feeding by these adults depleted the available MAM and apparently triggered weevil dispersal. In 2006 *R. latipes* was recovered 4 kilometers (2.5 miles) from the original release site and by the end of the 2007 season, *R. latipes* was recovered from, and had caused feeding damage to, MAM 5.6 kilometers (3.5 miles) from the release site.

# Other New Jersey releases

Between 2004 and 2007, PABIL released a total of 64,911 *R. latipes* adults into eight New Jersey counties (Table 4). Adult *R. latipes* and/or their feeding damage were observed in 2007 at eight of nine (88.9%) 2006 release sites. Overall, since 2004 the weevils have been recovered from 35 out of 37 (94.6%) of the sites (Table 4). That no weevils were recovered at two sites in Hunterdon County could have been due to rocky and dry conditions, and the fact that the weevils that were released were a mixture of new weevils and old weevils used for rearing. Also, because the releases took place in October the weevils may not have had sufficient time to acclimate before winter's onset.

Two of the release sites in New Jersey have been subjected to flooding. One of the 2005 Washington Crossing sites (Table 4) was located along the Delaware River and experienced a "100 year flood" in spring, 2006. The high waterline was two feet above the release site and no weevils were expected to survive; nevertheless, *R. latipes* adults were recovered at the release site in late May. Weevils were recovered from another site, along the Delaware River on the DOD Ponds Wildlife Management Areas in Salem County, even though it was periodically flooded by tides.

Adult *R. latipes* have dispersed from release sites in New Jersey, and some have migrated across the Delaware River to Amico Island. This ability to disperse is important because, despite weevil releases and other control activities, MAM is being found more frequently and over a greater range each year in New Jersey.

**Table 4**. Rhinoncomimus latipes releases and recoveries in New Jersey.

County	Location	Dates	Number Released	2008 Recovery
	2004 Releases	,		
Gloucester	Floodgate Road (shipped from the University of Delaware)	7/28	200	Y
	2005 Releases			
Salem	Abbott's Meadow WMA	4/22-9/9	6,976	Υ
Mercer	Washington Crossing State Park, near open air theater	5/13–5/18	652	Υ
Mercer	Washington Crossing State Park, near Delaware River	5/18	270	Υ
Gloucester	Floodgate Road	6/17–8/29	3,297	Υ
Salem	DOD Ponds WMA	10/12	600	Υ
2005 Total			11,795	
	2006 Releases			
Burlington	Pinelands Water Co.	4/28	2,260	Y
Burlington	Taylor's Farm	5/5	1,066	Y
Gloucester	Floodgate Road by Lake	5/12	800	Y
Mercer	Washington Crossing State Park	5/19	1,050	Υ
Salem	Supawna Meadows NWR for U.S. Fish & Wildlife Service	6/9-9/29	15,692	Y
Salem	Mesogianes Farm	9/1	1,125	Y
Hunterton	Hunterdon County Park	10/13	600	N
Hunterdon	Round Valley Rd	10/13	397	Y
Hunterdon	Pine Bank Rd	10/13	600	Y
2006 Total			22,465	

Table 4, continued. Rhinoncomimus latipes releases and recoveries in New Jersey.

County	Location	Dates	Number Released	2008 Recovery
	2007 Releases			
Salem	Lighthouse Road.	5/4	1,202	Y
Salem	Supawna Road 3 locations	5/4, 5/11	3,364	Y
Gloucester	Davidson Road	5/11	2,100	Y
Hunterdon	Dreabrook Road	5/18	1,800	Y
Salem	Killcohook spoils off Finns Point access road	5/25	4,513	Υ
Hunterdon	Railroad Avenue	5/18	600	Υ
Salem	Harris Road 5 locations	6/1	1,800	Y
Salem	Mesogianes Farm	6/1	827	Y
Middlesex	Rutgers Horticultural Farm	6/15	719	Y
Salem	Gant Farm	6/29	1,802	Y
Salem	Pennsville-Auburn Road	7/13	1,161	Υ
Salem	Pinyard Road	7/20	2,049	Y
Warren	Delaware Lake	7/27	1,200	Y
Salem	Fort Mott Road	8/3	1,200	Y
Salem	DOD Ponds WMA parking lot	8/9	671	Υ
Union	Watchung Reservation	8/16	1,894	Υ
Salem	Hook Road	8/24	1,998	Y
Bergen	Overpeck Preserve	8/30	741	Y
Salem	Park Avenue	9/5	351	Y
Hunterdon	Pine Bank Road (field collected from Floodgate site)	9/13	200	Y
Hunterdon	Pine Bank Road (lab weevils)	9/14	300	Y
Hunterdon	Route 29 (one year old weevils)	10/5	159	N
2007 Total			30,651	•
Total			64,911	

# Release of *Rhinoncomimus latipes* in other states

Weevils were released in Delaware in 2004 at two sites within White Clay Creek State Park (Table 5). Site 1 was a diverse site with a variety of other plant species, especially *Rubus* sp., and various trees and shrubs. Mile-minute weed was greatly suppressed within the first year at this site, and by 2008 very little MAM could be found except at the sunny, exposed edges. Site 2 was more of an open monoculture, and MAM persisted through 2008; however, by that same year weevils and damaged plants were abundant, *Rubus* was out-competing MAM in sunny areas, and Japanese stiltgrass (*Microstegium*) was out-competing it in the shade.

Table 5. Rhinoncomimus latipes releases and recoveries in states other than New Jersey.

State	County	Location	Dates	Number Released	2008 Recovery						
Delaware	1	2004 Re	leases								
	New Castle	White Clay Creek State Park (2 sites)	7/21–28	400	Υ						
		2006 Re	eleases								
	New Castle	Peterson Marsh	6/9	1,200	Υ						
		2007 Re	leases								
	New Castle	Pea Patch Island	6/13	1,070	Υ						
	New Castle	Peterson Marsh	7/3	900	Υ						
Maryland		2006 Re	leases								
	Cecil	Garrett Island	6/16	500	Υ						
		2007 Re	leases								
	Cecil	Garrett Island	600	Υ							
	Howard	Meadowbrook Park 7/12 500									
Pennsylv	ania	leases									
	Chester	Laurels, BVA-CREP, BVA-Wetland (3 sites)	6/9	1350	Y						
	York	Codorus State Park (2 sites), PSECU Bldg.	6/1–7/13	610	Y						
		2006 Re	leases		,						
	Franklin	Letterkenny Army Depot	5/4-6/2	1800	Y						
	Dauphin	Wildwood Lake Sanctuary	6/2	600	Y						
	Potter	Sinnemahoning State Park	7/27	600	Υ						
		2007 Re	leases								
	Montgomery	Pennypack Ecological Restoration Trust	5/23-7/9	3500	Υ						
	Dauphin	Wildwood Lake Sanctuary (2 sites) 6/14 300									
West Virg	jinia .	2005 Re	leases								
	Wood	Muskingum Isl., Ohio Rivers NWR (2 sites)	6/10	400	Y						
		2006 Re	leases								
	Wood	Neal Island, Ohio Rivers NWR	6/16	1000	Υ						

Weevils have been released on Muskingum Island in the Ohio River Islands National Wildlife Refuge in West Virginia. They have dispersed throughout the island's 100 acres, across the Ohio River onto the mainland on both sides, and over 5 miles upriver. Weevils were released on Neal Island in 2006 and have spread throughout its 105 acres. A third release was conducted on Wells Island in 2008. Additional releases have been conducted in Delaware, Maryland, and Pennsylvania (Table 5, above); however, it is too soon to assess the results of these releases.

# Release of *Rhinoncomimus latipes* in replicated release arrays in Pennsylvania

Weevil dispersal, population growth, and impact in the field were studied in three replicated release arrays in Chester County, Pennsylvania (Lake 2007). One array was located at the Brandywine Conservancy's Laurels Preserve and the other two at the Brandywine Valley Association (BVA) Myrick Conservation Center (BVA-CREP and BVA-Wetland sites). Each array consisted of a central release point surrounded by a total of 76 monitoring points: 60 points placed on concentric circles spaced 5 meters apart to a maximum distance of 25 meters (Fig. 28); eight points 1 meter from the release; and eight points approximately 2.5 meters from the release. On June 9, 2005, 450 weevils were released in the center of each array.

# Generalized Map of Release Arrays

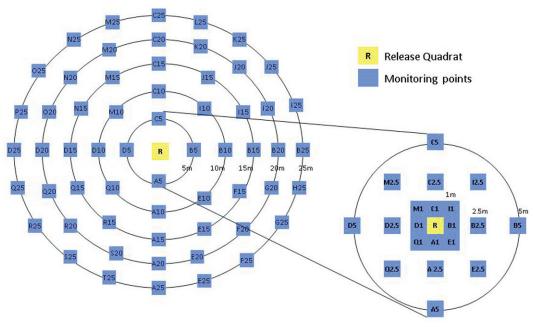


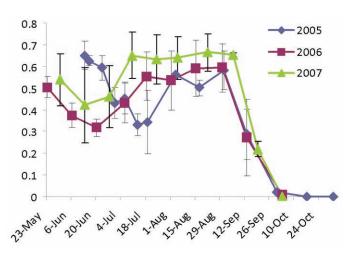
Figure 28. Generalized release and monitoring array for replicated releases.

# **Dispersal**

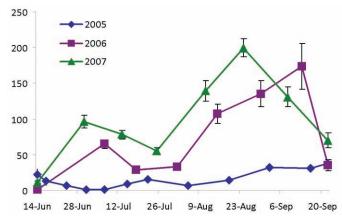
During the first three months following release, weevils had dispersed 15 to 25 meters within the arrays. Four months after release, long-distance dispersers were found up to 200 meters (0.12 miles) away. Within 14 months, weevils were found in MAM patches nearly 800 meters (0.5 miles) from the release. Dispersing weevils had located both large MAM populations and small isolated patches (Lake 2007). In 2007, approximately 27 months post release, weevils were found at several sites within 5.6 kilometers (3.5 miles) of the release points and at one site approximately 8 kilometers (5 miles) away. By June 2008, three years post release, weevils were observed on numerous MAM weed patches 11.3 kilometers (7 miles) from the original release sites. These patches ranged in size from small isolated vines to large infestations. As of July, 2008, the farthest-removed weevil dispersal was observed 29 kilometers (18 miles) from the nearest release sites.

## **Population growth**

Weevils were active in the field from early spring through fall and completed at least three or four generations. In 2005, 2006, and 2007, the proportion of monitored MAM weed quad-



**Figure 29**. Weevil egg production (proportion of sampled quadrats with eggs, average of all three sites each year).



**Figure 30**. Weevil population growth at the Laurels.

rats that contained eggs decreased from 60% in late August to zero in early October (Fig. 29). This decrease occurred before a substantial temperature drop, but coincident with a decrease in day length.

The percent cover of MAM varied greatly among monitoring points in the release arrays. In order to evaluate the weevil population in the context of different MAM cover, the number of weevils per monitoring quadrat was divided by the percent cover of MAM in that quadrat to generate the number of weevils per m² of MAM. For each site and year, the area under the curve for quadrats within 5 meters of the release was calculated based on the number of weevils per m² of MAM. At the Laurels it was found that weevil density increased significantly from 2005 to 2006 and from 2006 to 2007 (Fig. 30).

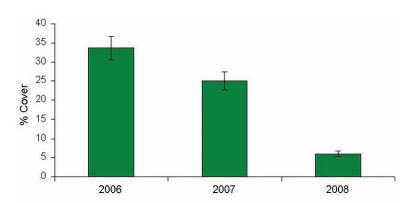
The average intrinsic rate of increase of the weevil population was estimated at 2.08 in 2005 and 5.03 in 2006. These rates are comparable to or exceed those of other successful biological control agents.

## **Impact**

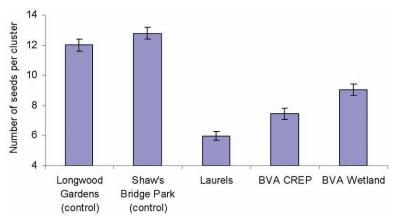
In addition to experiencing significant loss of photosynthate, damaged plants typically had stem nodes that were very close together ("stacked" nodes) (Fig. 31). The percent cover of MAM declined significantly between 2005 and 2007 at the Laurels (Fig. 32). The decline in percent cover at the BVA CREP site was close to significant and cover was unchanged at the BVA Wetland. The number of MAM seedlings per 0.5 m<sup>2</sup> declined significantly



**Figure 31**. "Stacked" nodes on weevil-damaged mile-a-minute plants.



**Figure 32.** Mile-a-minute weed percent of cover in early June at the Laurels.



**Figure 33**. Number of seeds per cluster at three release sites and two control sites.

between 2006 and 2008 at two of the three release arrays. At the Laurels—the array with the largest monoculture of MAM, as well as the largest weevil population—the number of seedlings declined from more than 100 seedlings per 0.5 m<sup>2</sup> in 2006 to fewer than 20 in 2008.

Weevil feeding significantly reduced the number of MAM seed clusters between 2005 and 2007 at two of the three release arrays. This damage also resulted in fewer seeds per cluster at the weevil release sites than at control sites with limited weevil activity. Two control (undamaged) sites averaged 12 seeds per cluster, while the Laurels had approximately 6 seeds per cluster (Fig. 33). The BVA sites had much smaller weevil populations than the Laurels had, but still had a significant reduction in the number of seeds per cluster compared to the controls.

#### Impact of R. latipes on MAM in field cages

The impact of *R. latipes* feeding on *P. perfoliata* was studied in field cages over a 2-year period (Hough-Goldstein et al. 2008; Fig. 34). In 2006, 20 weevils introduced into cages with single plants in May (when weevils first emerge from overwintering) suppressed

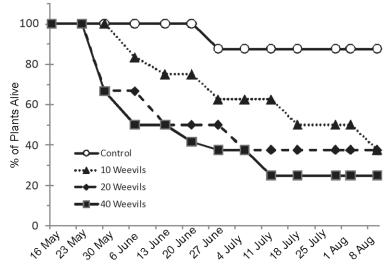


**Figure 34**. Mile-a-minute with heavy weevil damage in field cage.

seed production for about 9 weeks, whereas weevils introduced in June (when the first summer generation of adults emerge) did not affect seed phenology. Plants in all cages produced substantial numbers of seeds late in the year, but the average seed (achene) weight was reduced for plants with 20 weevils per plant introduced in May.

In 2007, plants grown within field cages, but with some competition from other plants, showed substantial mortality. By mid-August, 63% of plants with 10 or 20 weevils, and 75% of plants with 40 weevils per plant were dead, compared with 12.5% mortality for control plants (Hough-Goldstein et al. 2008) (Fig. 35). Reproduction was delayed by more than a month in surviving plants with 10 or 20 weevils, and by

more than 2 months in the few survivors with 40 weevils. Surviving plants with 40 weevils per plant showed loss of apical dominance, which can allow plants to compensate for herbivore damage; however, in the case of a light-adapted vine such as *P. perfoliata*, this may prevent the plants from achieving needed sun exposure. These results suggest that *R. latipes* feeding on *P. perfoliata* can impact plant growth and reproduction, and may put affected plants at a substantial competitive disadvantage.



**Figure 35.** Survival of individual mile-a-minute plants exposed to 0 (Control), 10, 20, or 40 weevils in field cages in 2007.

# Chapter 4. Biological Control as a Component of an Integrated Mile-a-Minute Weed Management Program

## **Integrated weed management**

Integrated weed management (IWM), an offshoot of the concept of integrated pest management that was applied first to insect pests and subsequently to plant disease pests, is rapidly gaining acceptance among weed scientists (Buhler 2002). Among the key elements of integrated pest management are the use of multiple control tactics and the integration of a thorough knowledge of pest biology into the management system. Elements of IWM systems may include:

Education and prevention

- Physical or mechanical control
- Cultural methods
- Herbicides
- Biological control

The ultimate goal of an effective weed management program in a natural area is to replace undesirable plants that cause resource, economic, habitat, or aesthetic losses with a plant or plants that are beneficial to the environment. The short-term objective is to implement the most effective combination of control methods available for the target weed. Concurrently, landowners and managers should develop a long-term plan for managing undesirable plants and maintaining desirable vegetation.

# Weed control methods used to manage MAM

#### **Education and Prevention**

Because mile-a-minute weed is still expanding its range (Fig. 2, page 2), and is patchily distributed even in areas where it is well entrenched, efforts to increase public awareness of this noxious weed are important to the success of any area-wide integrated management program. Mile-a-minute weed can grow to unmanageable proportions within a fairly short time of establishing itself in a new area. For example, the plant was first noticed in very small patches in 2001 in the heronry on Pea Patch Island, Delaware. The extent of infestation was mapped in 2002, when the population was still small, and in 2003 and 2004, when populations exploded (Fig. 36). Although not mapped, populations on the remainder of the island outside of the heronry increased in a similar pat-



Figure 36. Mile-a-minute distribution in heronry, Pea Patch Island, Delaware.



Figure 37. Mile-a-minute on Pea Patch Island, Delaware, August 2003.

tern (Fig. 37). Populations of MAM remained extremely high from 2005 through 2008. Weevils were released in 2007 and 2008, and their progress is being monitored.

MAM can respond rapidly to disturbance. For example, in 2003 MAM was probably present on a Chester County, Pennsylvania, site slated for development, but the weed was not a problem at that time. In the spring of 2004, soil disturbance occurred during testing to determine septic feasibility. By the spring of 2008, the site was a virtual monoculture of MAM, with an average of 86 seedlings per square meter (E. Lake, unpublished data).

In areas where MAM is present, land managers must anticipate the potential

for it to colonize and/or dominate disturbed sites. It can also dominate land cleared for restoration projects. For example, a preserve in Chester County, Pennsylvania, decided to convert a site with a mixture of woody and herbaceous invasives, including MAM, to a native meadow. Heavy equipment and herbicides were used to prepare the site in the fall of 2007, and a mixture of native grasses and wildflowers was seeded. The following spring, the site was a monoculture of MAM with little to none of the desirable vegetation visible.

Eradication may be possible where a population is still small. For example, a nursery in Kingston, Rhode Island, has successfully controlled a small infestation through hand pulling and mowing, though an occasional plant still recurs and is removed (R. Casagrande, Univ. Rhode Island, personal communication).

#### Physical or mechanical control methods

Mile-a-minute weed has a relatively weak root system, and small plants can be hand-pulled easily. Gloves should be worn to protect the skin from the plant's sharp spines. Longer vines can be pulled out using a garden rake, as has been done in parts of Little Paint Branch Park, near Beltsville, Maryland (Marc Imlay, personal communication). Regardless of the method used, to avoid spreading seed to new locations weeds should be pulled before they set seed.

Even green seed can germinate (see "Germination of mature and immature seed"); therefore, if any seed clusters are present, plants should be removed from the area and the seed destroyed. Adequate methods of destroying MAM seed have not been confirmed through research, but experiments on other types of weed seeds suggest possible

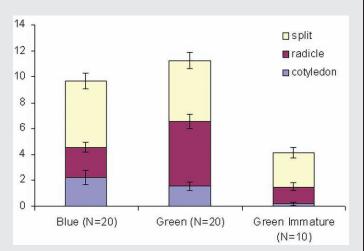
#### Germination of mature and immature seed

A common reaction of land managers to the appearance of seed clusters on an uncontrolled MAM infestation is to attempt to remove the vines or apply post-emergent herbicides (personal observation). It was not known whether viable green seed was present at the time these management techniques were implemented, and if this management strategy could further the spread of MAM and increase the seed bank.

Blue, green (full sized but green) and green immature (green and not full sized) fruits were collected from MAM vines in the fall of 2004. The fruits were dried and the perianth was removed leaving only the achenes. Achenes were separated into groups of twenty, put into Ziploc<sup>®</sup> plastic bags with moist peat moss and placed in a refrigerator for cold stratification on June 29, 2005. After approximately 7 weeks, seeds were removed from the refrigerator and the bags were placed in a greenhouse (Colpetzer and Hough-Goldstein 2004). One week later the seeds were removed from the peat moss

and checked for germination. The seeds were categorized as: no sign of germination, seed split, radicle present or cotyledon present.

A proportion of green MAM seed was shown to be viable (Fig. 38); therefore, removal and/or movement of vines with green seed could contribute to the spread of MAM and add viable seed to the seed bank. Managers should take the potential viability of green seed and the annual variation in the timing of seed development into consideration when implementing management techniques.



**Figure 38**. Mean number of blue, green, and green immature seeds that germinated after cold stratification (20 seeds per sample).

methods. For example, high temperatures in an active compost pile can destroy weed seed, but internal temperatures need to reach at least 60° C (140° F) for 7 days to kill weed seeds (Rynk 1992). It is very difficult to reach these temperatures near the surface of compost piles, so only weed seeds in the interior of the pile are killed (Gordon et al. 2001). Therefore, to ensure all seeds are exposed to the internal temperatures, compost piles need to be turned or mixed periodically. Burning can kill seeds, but the fire must be very hot. Work in Australia suggests that 400 °C (750°F) for 20 to 30 seconds may be required for hard-coated seeds (Walsh and Newman 2006), so simply putting vines in a trash barrel and burning them may not be adequate. Ultimately, until more effective seed-killing methods are determined, land managers should make every effort to control the plant before seed clusters develop.

Where practical, mile-a-minute weed can be mowed. However, while low mowing may kill plants, leaving too much of the plant above ground can release apical dominance (Fig. 39) and cause re-growth of sturdy bushy plants, possibly with more terminals and a consequent increase in the potential to produce more seed clusters.



Figure 39. Side terminals produced by mile-aminute plants that have been mowed.

As noted earlier, MAM seed can persist and remain viable for 6 years in the seed bank. Even if plants are removed or killed by physical means, the efforts must continue for several years to exhaust any remaining seed bank.

#### **Cultural methods**

Observations and experiments suggest that MAM does not thrive in the shade; therefore, one important component for controlling it long-term is to add shade trees wherever practical and desirable. Competition with other plants is also key in determining whether MAM can dominate a site. Fostering desirable plants, whether by planting or relying on natural populations, should be part of a management plan (Fig. 40).



Figure 40. Damaged mile-a-minute plant, with competing native vegetation.

#### **Herbicides**

Kevin Fryberger, Preserve Manager, Brandywine Conservancy, Chadds Ford, PA, and Art Gover, Research Support Associate, Dept. of Horticulture, Penn State University, College Park, PA.

Mile-a-minute weed is quite susceptible to a number of herbicides, including both pre- and post-emergent products. **Pre-emergent herbicides** control plants before they emerge from the ground by injuring the plant as the seed germinates. They can be applied to soil and will enter the plant through the roots or shoots emerging from the germinating seed. **Post-emergent herbicides** act on plants after they have emerged from the ground, entering the plant through the foliage or stems. Some herbicides work both ways.

Effective control using herbicides is easiest early in the season, before seed set and before MAM begins to climb onto neighboring plants. Depending on weather, seasonal conditions, and the herbicide you choose, pre-emergent herbicides should be applied between early March and early April. An advantage of pre-emergent applications is that most of the existing perennial species will not be affected, particularly if you apply prior to bud break.

Post-emergent herbicides may be selective or non-selective. Glyphosate is a widely used non-selective herbicide. When used on very dense stands or monocultures of MAM, it will provide effective control with minimal damage to non-target plants. However, when MAM is growing among desirable plants, it may be very difficult or tedious to avoid contacting the non-target plants. If most of the desirable plants are grasses or grass-like, a selective herbicide such as triclopyr ('Garlon® 3A') can be useful. Triclopyr will control MAM without injuring grasses, but will injure on contact other broadleaf forbs, shrubs, and trees. Other effective post-emergent products include Journey®, Plateau®, Overdrive®, Escort®, and Milestone® VM.

Adding a **surfactant** is recommended for better control of MAM with post-emergent sprays. Surfacants improve herbicide effectiveness by increasing the spray's adherence to the leaf surface, reducing the surface tension of the mixture so that it spreads over more of the leaf, and aiding penetration of the waxy outer cuticle of the leaf, all of which promotes better uptake of herbicide into the treated leaf.

Several herbicide products are readily available for both consumer and commercial applicators. Generally, consumer products are less concentrated, and come in smaller containers than commercial products. Any of the commercial products listed in Table 6, below, could be purchased for home use: none of them are "Restricted Use" meaning the purchaser need not be a state-certified pesticide applicator. However, if you buy a commercial product for residential use you will likely end up purchasing much more material than you will need. Although the unit cost of commercial products is lower, the larger container size can make them too expensive for small-scale use. Plus, you must store or dispose of the surplus product, safely.

Commonly used pre-emergent (PRE) and post-emergent (POST) herbicides are listed in Table 6, below. Pre-emergent herbicides commonly used to control MAM are not readily available as consumer products. The herbicide pendimethalin is available in some crabgrass-prevention products, but often includes fertilizer and is intended for use on established turfgrass.

Prior to applying any herbicide, please check your equipment thoroughly and consult the product label for proper application rates and precautions.

Table 6. Commonly used herbicides for mile-a-minute control.

Herbicide	Pre or post emergent	Homeowner product name and concentration	Commercial product name and concentration
pendimethalin	Pre	Halts® Crabgrass Preventer (1.7 %)	Pendulum® Aquacap™ 39% Pendulum® 3.3 EC 37% Pendulum® 2G 2%
imazapic	Both	None	Plateau® (Govt. only) 24% Journey® (plus glyphosate) 8%
sulfometuron	Both	None	Oust® XP 60%
glyphosate	Post	Many 1% Many 18% Many 41%	Roundup® Pro 41% Rodeo® 54%
triclopyr	Post	Roundup® Poison Ivy & Tough Brush Killer 8%	Garlon® 3A 44% Garlon® 4 62%

<sup>&</sup>lt;sup>a</sup> Brand names are listed for reference only. All herbicides listed are available in other products as well. *Glyphosate* is so widely available that homeowner product examples are listed by common concentrations rather than brand

There are many factors to consider when selecting an herbicide, including time of year, surrounding vegetation, rate of infestation, herbicide volatility and translocation. Factors that can cause variation in results include rainfall during or immediately after application, and drought. Drought-stressed plants are usually less responsive to herbicide applications than are actively growing plants.

The ability of a restoration site to recover from weed competition once the weeds have been removed will determine short- and long-term management decisions. Complete control may not be feasible. The most efficient and effective strategy results from a thorough understanding of the environmental forces in the area and a management goal that works with and not against these forces. There are many techniques for controlling MAM. Usually, the control on a site will require a combination of two or more methods. What will be common to every site is that, owing to the prolific nature of MAM and the persistence of the seed bank, periodic monitoring over many years will be required to prevent a disruption to the aesthetic and ecology of a site.

## The biological control component

#### Planning your program

Rhinoncomimus latipes has only been available for release against MAM in the U.S. for a few years; therefore, we have limited information concerning the weevils' success and population growth under various conditions. We need much more experience and research to determine optimum release rates and methods.

In areas where the MAM population is a massive monoculture that must be controlled quickly, such as where trees have been planted and are in danger of being overrun, it is probably wise to plan multiple modes of attack. Such a plan would include the application of pre-emergent herbicide in areas where other valued annual plants are not likely to be harmed; fostering or planting desirable plant species as competitors; and releasing weevils, which over time should increase their populations to the point where they will permanently suppress the target plant and help promote a healthy, diverse ecosystem.

#### **Selecting release sites**

Rhinoncomimus latipes are present and abundant on MAM throughout China, from north to south, so there is no obvious reason why weevil populations should not establish and develop throughout the current and future MAM range in North America. So far this has been the case. Weevils have established populations at nearly all sites where they have been released. In the mid-Atlantic region they can develop three to four overlapping generations over a single season (Lake 2007). Because of the cooler temperatures in more northerly regions, we would expect fewer generations, and therefore it may take more time for large populations to develop further north.

After release and while the insect populations are developing, at least a portion of the selected release site should remain undisturbed by other methods of control, e.g., herbicides, mechanical methods, etc. The site selected should be one where the MAM population can be tolerated for several years.

### Obtaining weevils for biological control

R. latipes will be commercially available from the Phillip Alampi Beneficial Insect Laboratory (PABIL), New Jersey Department of Agriculature, in April, 2009. In states where weevils have been established, weevils can be moved legally from one site to another within the state. As weevil populations increase, cooperative extension agents may hold "field days" at sites where large numbers of weevils are produced, encouraging homeowners and landowners with MAM infestations to come and collect weevils for release at their own sites.

Weevils can be reared if resources are available (see Chapter 3, Mass Rearing); however, they can be shipped or transported across state lines only if a USDA-APHIS-PPQ 526 permit is obtained in advance. The form for requesting this permit, along with other relevant information, is available online, at http://www.aphis.usda.gov/plant\_health/permits/organism/index.shtml.

#### **Monitoring weevils**

In order to document effects of biological control on the target plant population, it is important to keep accurate records of when, where, and how many biocontrol insects are released, and track the population of the insects and their subsequent impact on both the target weed and the entire plant community. Ideally, to make sure that any observed changes are not due simply to varying or seasonal conditions, or that they would have occurred with or without the introduced insect, one would keep track of several weed populations that are exposed to the insect and other, similar weed populations not exposed to the insect. That said, if an introduced insect is a successful biological control agent, sooner or later its population is likely to increase to the point that any control site will be invaded by the insect, at which time the site will cease to function as a control.

The MAM monitoring protocol has gone through several iterations as we have gained experience with the plant and the weevil. The initial protocol was based on monitoring protocols for purple loosestrife and garlic mustard developed by Bernd Blossey, Victoria Nuzzo, and coworkers (http://www.invasiveplants.net/). The 2008 version of the MAM monitoring protocol is included here as Appendix A, and the latest version is available online at http://ag.udel.edu/enwc/research/biocontrol/mileaminute.htm.

The MAM monitoring protocol is designed to track the population of the weevil and the MAM population over time. Ten permanent 0.5- by 1.0-meter quadrats, numbered 1-10, are established in a heavily infested MAM patch where weevils are to be released, and ten quadrats are established in a similar control site approximately 500 meters away. Weevils are then released in quadrat #5 of the first array. The full monitoring protocol calls for a spring sample, where MAM seedlings and weevils are counted within a quadrat frame (Fig. 41). Once a month following the spring sample, weevils observed within quadrats are counted, the percentage of leaf area removed by insects is estimated, and presence or absence of node damage indicating weevil reproduction is noted. Later in the season the number of mature and immature fruiting terminals is counted. The percentage of MAM cover in each quadrat is estimated during each survey. The expectation

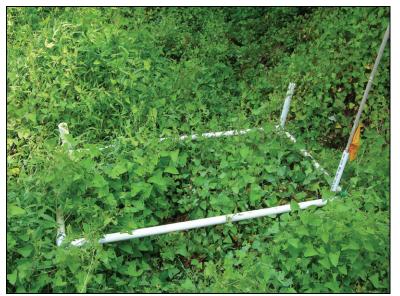


Figure 41. Frame used for monitoring mile-a-minute and weevils.

is that as the weevil population increases, the percentage of MAM cover will be reduced in the quadrats.

Recognizing that not all land managers who release weevils have the labor or time resources to follow the complete monitoring protocol, we have developed an alternative "quick" protocol (Appendix B). This protocol involves physically marking the site where weevils have been released, and going back three times per year (spring, summer, and fall) to observe whether weevils, weevil feeding damage, and node damage can be observed within a 1-meter radius of the release site. In addition, the approximate percentage of MAM and other plant coverage is estimated at each site. The sites should be

revisited for at least 3 to 5 years, or until the MAM has been suppressed. In addition, a digital photo of the release site should be taken at the same time every year (late summer or early fall, before frost) for a visual record of changes in vegetation.

#### **Combining biological control with other methods**

Additional research is needed (and underway) to help us determine which MAM control methods are most compatible with the weevils. Although herbicides are not likely to have a direct detrimental effect on adult weevils, death of the plant will likely kill any developing larvae in the stems, and cause adults to disperse. High mowing of sites with weevils may make the site more conducive to weevil population growth by causing the plants to produce more of the tender terminals favored by the weevils, but this has not yet been proven.

As noted, competition with other plants is key in determining whether MAM can dominate a site. Weevil damage can cause MAM plants to become poorer competitors by reducing the number and size of seeds, shifting phenology of seed production to later in the year, and suppressing plant growth (Hough-Goldstein et al. 2008).

An important aspect of developing an integrated weed program is to assess the other vegetation that is present at a site dominated by MAM. Not much is gained if biological control agents suppress the target weed, only to have the target weed replaced by other nonnative invasive plant species. In some cases, control of MAM by whatever means should be followed or accompanied by planting of desirable vegetation.

# **Conclusion**

The biological control program for *P. perfoliata* in the eastern U.S. was initiated in 1996, and a permit for release of the host-specific weevil, *R. latipes*, was obtained eight years later, in 2004. Although much remains to be learned, our first 4 years of experience suggest that this weevil will be very successful overall in suppressing the target weed. The weevil shows all the characteristics of a desirable biological control agent, including: a high reproductive rate, with three to four overlapping generations occurring each season; extreme host specificity; excellent dispersal capabilities; and the ability to suppress the target weed. Time will tell the extent to which it will control mile-a-minute weed in a variety of environments throughout the introduced range.

#### **APPENDICES**

# Appendix A: Mile-a-minute weed monitoring protocol Revised, March 2008

Judy Hough-Goldstein, Department of Entomology & Wildlife Ecology, University of Delaware (jhough@udel.edu)

#### Introduction

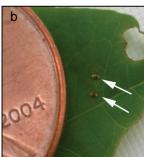
Mile-a-minute weed (*Persicaria perfoliata*) is an annual Asian vine that invades a variety of habitats in the northeastern United States, including forested floodplains, streamside herbaceous wetlands, and upland forests. A biological control program targeting mile-a-minute weed (MAM) was initiated by the Forest Service in 1996, with field surveys and laboratory host-specificity tests conducted in China and subsequent testing under quarantine conditions in Delaware. A stem-boring weevil, *Rhinoncomimus latipes* Korotyaev, has been determined to be host-specific to MAM and a permit application for field release was approved in July 2004. The following guidelines are intended to help monitor the abundance of both MAM and the weevil, and assess the long-term impact of biological control. Ideally, monitoring should be initiated one or more years before biological control organisms are released, so that changes can be tracked pre- and post-release.

Mile-a-minute weed is a prickly, branching, viney annual plant that germinates in early spring, usually in April in the mid-Atlantic region. Vines grow rapidly, climbing over other plants, and attain lengths of 6 m or more. Flowers are inconspicuous, and iridescent blue berry-like achenes are produced, beginning in mid-summer and continuing until the plants are killed by frost in the fall. Seeds require a cold period before germinating. Many will germinate underneath established patches the following year, while others are spread by birds, mammals, water, and in the treads of shoes and tires. Mile-a-minute seeds can survive for up to 6 years in the seed bank.

Adult *R. latipes* are about 2 mm long, and are black, but once they start feeding they may be covered by an orange film derived from plant exudates. Adult weevils eat small holes in young leaves of *P. perfoliata* and lay eggs on leaves and stems. After hatching, larvae bore into the stem where they complete development, then exit the stem and drop to the soil for pupation. Development from egg to adult takes about 26 days under laboratory conditions. Weevils are very small, but can be observed directly in the field, especially at the ends of ter-

minals (Fig. 42a). The pale yellow eggs have a characteristic peanut shape and are covered by a thin strip of fecal material (Fig. 42b); however they are difficult to spot in the field due to their very small size. Characteristic adult feeding holes ("shot holes" in leaves) are relatively easy to see (Fig. 42c). Larval emergence holes at plant nodes (near where ocreae encircle stems or where stems diverge) can sometimes be seen in the field (Fig. 42d).





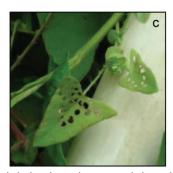




Figure 42. a) Adult weevil; b) eggs with penny; c) adult feeding damage; d) larval node damage.

Because of MAM's rampant summer growth, and the fact that all reproduction is by seed (since plants die off, roots and all, following one or more hard frosts in the fall), we believe that plant populations are best assessed in the spring, when individual small plants can be counted in measured quadrats. Counts of overwintered weevils can be done at this time, too. Summer/fall assessments of weevil abundance and damage should be conducted, too, because populations will increase and spread during the summer. Summer and fall counts of seed production will help assess impacts on the plant population.

# Site selection and quadrat placement

#### Selection

Select a weevil release/monitoring site that will be protected from other uses that could jeopardize insect establishment and continued monitoring, i.e. a site where the landowner will not attempt to control vegetation through mowing, herbicide use, etc. The study site should contain an ample population of MAM; however, ideally, native vegetation should be present so that control of MAM will result in the establishment of a more-desirable plant community.

#### **Quadrat placement**

#### Materials needed

One 0.5 m² quadrat frame (see "Constructing a quadrat frame," below), 80 pieces of 0.5-inch or 0.75-inch plastic conduit pipe ~1 m long (to mark corners of 20 quadrats in each site), hammer, permanent marker, 50 or 100-m tape measure, GPS unit (if available), camera, work gloves

Within a single monitoring area (e.g. a state park), establish two 100-m-long transects, with similar habitat, vegetation, and mile-a-minute populations, but located approximately 500 m away from each other. Randomly assign one of these transects as the "release" transect, where weevils will eventually be released. The other transect will serve as

# **Constructing a quadrat frame**

#### **Materials**

One 10-foot length of 0.5- or 0.75-inch-diameter PVC or CPVC conduit pipe; four right-angle elbows of the same diameter; PVC or CPVC glue; hacksaw or pipe cutter; permanent marker; measuring stick or tape measure.

# Do not glue assembly a to assembly b

**Figure 43**. Diagram of a quadrat frame.

#### **Assembly**

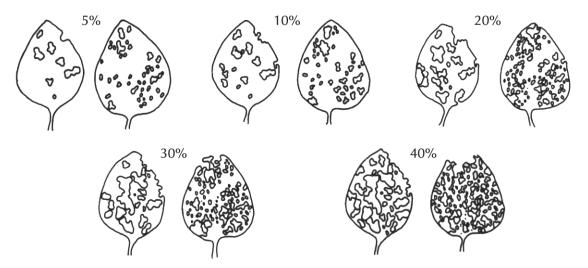
The inside dimensions of the finished frame should measure 1 m by 0.5 m.

Using the hacksaw or pipe cutter, cut the pipe into four pieces; two pieces 1 m long, and two pieces 0.5 m long.

Glue an elbow to each end of one of the long pieces (a), taking care that the elbows are perfectly aligned with each other (share the same right-angle plane). Set this assembled piece aside; it will be the fourth side of the frame. (b) Glue the elbows on the remaining long piece and then glue a short piece into each elbow so as to form an open U-shaped frame. Using a permanent marker, mark 10-cm intervals on each side to assist in estimating percent cover and seedling numbers.

a non-release control site, at least until the weevils disperse into it. Along each transect, locate 10 quadrats, approximately 10 m apart. Permanently mark the position of each quadrat by placing the quadrat frame (Fig. 43) on the ground and hammering a 1-m long piece of conduit pipe into the ground at each inside corner. Using a permanent marker, write the quadrat number (R-1 through R-10 for the release transect and C-1 through C-10 for the control transect) on each corner pipe. Remove the frame but leave the pipes in the ground as markers for future reference. Move 10 meters along the transect and repeat the process until you have ten sets of quadrat markers in the ground. If 100-m long patches of MAM are not available, the 10 quadrats can be placed at random within the MAM infestation. Quadrat #5 will serve as the release point and should be located near the center. Make note of the approximate distance between quadrats on a sketch or map and attach it to Form 1 (below), along with GPS coordinates and/or landmarks to help to find the quadrats, later. A brightly colored flag placed in one of the corner pipes will also help when locating a quadrat. Identify permanent photo-points and take photographs of the study site, including one or more set of markers. Leave the markers in place until you've completed the study. Note: be sure to remove all the markers when you complete the study.

Establish the quadrats initially during the period of MAM germination and seedling emergence, making sure each quadrat has a MAM population. (Note, if other tearthumbs are present you may need to wait until plants have developed characteristic ocreae, encircling stems, before establishing quadrats).



**Figure 44**. Percent of defoliation. Note, examples of damage up to 40% are shown, but damage up to 100% can be estimated.

# Weevil release

Release approximately 200 or more adult weevils within quadrat #R-5. Carefully document all releases, including the date, numbers released and exact site of release.

# **Spring mile-a-minute survey (Form 1)**

#### Materials needed

Ruler, Form 1 (make copies as needed), clipboard, pencils, camera, maps, work gloves, GPS unit, hand tally.)

Choose a date in spring after the main flush of mile-a-minute germination is complete (probably between April 15 and May 15), but before vines have become too dense to count. Ideally, MAM stems should be 15-30 cm (6"-12") tall. Measure the height of an average stem (or a range of heights if there is much variation) and note it on the survey

We maintain a data base of all North American releases, so please inform our lab of any new releases as soon as they occur. Please send the following information to Dr. Judith Hough-Goldstein at jhough@udel.edu:

- Date of release
- Number released
- State
- County
- Location (park, refuge, or municipality)
- Description of exact site
- GPS coordinates, if available

Also, please let us know where you obtained the weevils (e.g. Philip Alampi Laboratory, or from an established field site), and send us copies of all monitoring forms (see below) at the end of the season.

form. Slide the quadrat frame in place around the four corners, and survey each quadrat for the following:

 Number of weevils (if released in previous years, or where weevils may have spread on their own). Adult weevils tend to drop from plants when disturbed, so approach each quadrat site carefully. First count and record all adult weevils that can be seen on plants within the approximate confines of the quadrat. Weevils will generally be on MAM terminals or foliage, often near characteristic "shot hole" feeding damage.

- Total number of mile-a-minute seedlings present in the quadrat. Use a tally counter for accuracy. If too many are present to count, you may count the number in half or a quarter of the quadrat and multiply by 2 or 4 to arrive at a reasonable estimate within the entire quadrat; however, if you do this, to avoid errors note it on Form 1 under "Comments" (last column).
- Number of stems of other plant species. Identify as many other species as possible, especially those that are most abundant, and note these on the form under "Comments."
- Percent cover. Standing over the frame, look straight down and estimate how much of the
  quadrat is covered by mile-a-minute foliage and vines, and how much is covered by all
  other vegetation (these estimates may total >100%, due to layering).
- Note presence or absence of "shot holes", the characteristic damage of feeding weevils.

# Summer/fall assessment of weevil abundance, plant damage, and seed production

Materials needed: Form 2 (make copies as needed), clipboard, pencils, work gloves, (optional, GPS unit, hand tally). Once each month following the seedling counts (or weevil release) until plants senesce, return to each quadrat site and survey for the following within each quadrat and record your findings on Form 2 (below) (Note: if substantial mile-a-minute growth has occurred, it may be necessary to search out, locate, and cut a path to, each quadrat the week before the samples are taken):

- Number of weevils. Carefully approach each quadrat site and first count and record all adult weevils that can be seen on plants within the approximate confines of the quadrat.
- Percent defoliation. Scan the foliage for "shot holes" in leaves, the characteristic damage caused by feeding weevils, and assess the percentage of leaf area removed from mile-a-minute foliage within the approximate confines of the quadrat (see Fig. 44, above). If insects other than the weevil are present, e.g., Japanese beetles, and appear to be damaging the foliage note this under "Comments" on Form 2.
- Node damage (yes or no). Look closely at stems where adult weevil feeding damage is evident, and note presence or absence of node damage, where larvae have fed in stems or emerged for pupation.
- Percent mile-a-minute cover. Standing over the frame, look straight down and estimate how much of the quadrat is covered by green (not senescent) mile-a-minute foliage and vines.
- Number of fruiting terminals. Once seed clusters have formed, count the number of mature (containing at least one blue or purple seed) and immature (all green) seed clusters within each quadrat.

Form 1. Spring mile-a-minute survey.

To be conducted after flush of MAM germination, probably between April 15 and May 15, but before vines become too dense, when stems are between 15 and 30 cm (6 to 12") long.

Site

Approximate length of MAM stems Person(s) conducting survey\_\_\_\_\_

This form is available as a Microsoft @Excel® spreadsheet. Contact Dr. Judith Hough-Goldstein, jhough@udel.edu.

Comments (Other plants present in the quadrat, etc.)																					
Weevil feeding damage	8																				
We fee dan	Yes																				
Percent cover in quadrat	Other																				
	MAM																				
nber of stems iginating in quadrat	Other																				
Number origina qua	MAM																				
Number of Number of stems weevils originating in (count first)																					
Quadrat number		R-1	R-2	R-3	R-4	R-5	R-6	R-7	R-8	R-9	R10	C-1	C-2	C-3	C-4	C-5	O-6	C-7	C-8	C-9	C-10

Date\_

Form 2. Summer/fall weevil and plant monitoring.

To be conducted monthly.

Date\_\_\_\_\_

Person(s) conducting survey\_\_\_\_\_

Comments (Other plants present in the quadrat, etc.)																					
oer of ing nals*	Immat.																				
Number of fruiting terminals*	Mature Immat.																				
Percent MAM cover in quadrat																					
Node amage	9 N																				
ס	Yes																				
Percent of leaf area removed by insects																					
Number of weevils (count first)																					
Quadrat number		R-1	R-2	R-3	R-4	R-5	R-6	R-7	R-8	R-9	R10	C-1	C-2	C-3	C-4	C-5	C-6	C-7	8-0	6-O	C-10

\*Mature fruiting terminals have at least one blue/purple seed; immature terminals have buds or seeds that are all green.

# Appendix B: Mile-a-minute weed quick monitoring protocol, July 2008

Judy Hough-Goldstein, Dept. Entomology & Wildlife Ecology, University of Delaware (jhough@udel.edu)

Not all land managers have the labor or time to follow the complete monitoring protocol. Therefore, we developed an alternative "quick" protocol:

- Physically mark the location where weevils are released, using a wood, metal or plastic stake with flagging tape.
- Determine the latitude and longitude of the release point using GPS or Google® Earth.
- Record the date and number of weevils that are released, along with a descriptive label (e.g. Longwood-1) and detailed description of the location so it can be found again even if the physical marker is lost.
- Estimate the percent cover of mile-a-minute (MAM) in the area immediately surrounding the release point (within a circle of ~1-meter radius), to the nearest 5%.
- Note what other plants are dominant within that circle.
- Take a digital photo of the release site and return every year at the same date (late summer or early fall, before frost) to take another photo from the same location (vantage point). Include the release plot and surrounding area in the photo. Note the date and if possible include a noticeable landmark in the photo.

Go back to the release point three times each year (Spring, Summer, and Fall), and record the following:

- Date, location.
- Presence and extent of weevil feeding damage on MAM (none, low, medium, or high damage; see form for description of categories).
- Number of weevils (if any) found within ~1-meter radius of the release point; search and count for at least 2 minutes.
- Presence of node damage on MAM, indicating egg production and larval tunneling.

- Estimated percent cover of MAM in the area immediately surrounding the release point (within a circle of ~1-meter radius), to the nearest 5%.
- Other plants that are present within that circle.
- If MAM and/or weevils or damage are not present within the immediate area of the release, search the surrounding area and note the presence or absence and extent of MAM and weevils or feeding damage.

Images of MAM, weevils, feeding and node damage can be found at

http://ag.udel.edu/enwc/research/biocontrol/index.htm

Please record your results on the survey form (Form 3, below) and send it to:

Dr. Judy Hough-Goldstein
Dept. of Entomology & Wildlife Ecology
Townsend Hall Rm. 250
531 South College Ave.
Newark DE 19716-2160
jhough@udel.edu
302-831-8889 fax

BIOLOGY AND BIOLOGICAL CONTROL OF MILE-A-MINUTE WEED	
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Form 3: Mile-a-Minute Quick Monitoring Survey

# **Mile-a-Minute Quick Monitoring Survey**

(Survey to be conducted at time of weevil release and three times per year thereafter, once each in spring, summer, and fall, until MAM is suppressed [3 – 5 years])

Site location (descriptive label):
Date:
Person(s) conducting survey:
Phone # or e-mail of person conducting survey or supervisor:
At time of release:
Physically mark the site of release, and attach a map/sketch and description of the release site, with enough detail that the site can be found again even if the physical marker is lost.
Site latitude and longitude:
Number of weevils released:
Estimated % cover of MAM (to nearest 5%) within ~1-meter radius of release:
Other plants dominant within ~1-meter radius of release:
When site is revisited:
Estimated % cover of MAM (to nearest 5%) within ~1 m radius of release:
Weevil feeding damage on MAM at release site (circle one): none, low, medium, high
(low = holes in a few scattered leaves; medium = holes in many leaves; high = extensive damage on most leaves)
Number of weevils within ~1-meter radius of release point (2-minute [minimum] count):
(Note: Typically, counts are low in the year of release and the following year.)
Visible node damage on MAM within ~1-meter radius of release point (yes or no):
Other plants dominant within ~1-meter radius of release point:
If MAM and/or weevils are not present within ~1-meter radius of release point, search the surrounding area and note presence and extent of MAM, weevils, and weevil feeding damage:
Comments:

#### **G**LOSSARY

abdomen The last of the three insect body regions; usually contains

the digestive and reproductive organs.

achene A small, dry, indehiscent fruit with a single seed.

alternate Leaves that are arranged singly at each node along a stem.

annual A plant that flowers and dies within a period of one year

from germination.

apical dominance Influence exerted by a terminal bud in suppressing the

growth of lateral buds.

aspirator An apparatus used to suck insects into a collection con-

tainer.

beetle A member of the very large and variable insect order

Coleoptera; adults have hardened or leathery forewings (elytra) while larvae may be grub-like or mobile; beetles

exhibit complete metamorphosis.

biological control The reduction in the abundance of a pest through inten-

tional use of its natural enemies (predators, parasitoids,

and pathogens); also called "biocontrol."

biological control agent A natural enemy of a target pest used in biological con-

trol efforts.

chemical weed control Weed control strategies employing herbicides.

choice test A test of host specificity in which the potential biologi-

cal control agents are presented with a combination of test-plant species along with the target weed, and their

oviposition or feeding is recorded.

classical biological control A biological control strategy employing the release of

a pest's natural enemies imported from another region; typically directed against exotic pests, it uses natural

enemies from areas where the pest is native.

cold stratification A period of moist cold required for some seeds before

they will germinate.

competition Negative interactions between individuals of the same

or different species that utilize the same resource(s); if the resource is in short supply, one individual or species may survive and increase in number at the expense of the

other(s).

complete metamorphosis A type of insect development characterized by immature

stages (larvae and pupae) that look quite different from the adults, and typically live in different habitats, eat different foods, and exhibit different behaviors than do the

adults.

community A naturally occurring group of different species of organ-

isms that live together and interact as a more or less self-

contained "unit."

cover The portion of the vegetative canopy in a fixed area at-

tributable to an individual or a single plant species.

cultural methods Weed control methods that modify the plant's environ-

ment, such as adding or removing shade or fostering

competition with other plants

defoliation The loss of foliage, often due to insect feeding.

defoliator An organism, usually an insect, that consumes plant foli-

age.

density Number of individuals per unit area.

dispersal The spread of animals and plants from any point; the

redistribution of plant seeds, fungal spores, or insect eggs,

larvae, and adults.

dormant In a state of temporarily reduced metabolic activity.

emergence Act of adult insect leaving the pupal case or reappearing

after overwintering.

eradicate Total elimination of an organism from an area.

excelsior Long, thin wood shavings used for packing.

exoskeleton A hard outer structure, such as the shell of an insect or

crustacean, that provides protection or support for the

organism.

exotic Not native.

FHTET Forest Health Technology Enterprise Team, a division of

the USDA Forest Service

field insectary

An area where host plants or animals are abundant and

biological control agents are released and propagated with or without additional human manipulation.

forb A herbaceous plant that is not a grass nor grass-like in

form.

frass The excrement produced by insects, containing feces and

undigested plant material

genus (pl. genera) A taxonomic category ranking below a family and above

a species and generally consisting of a group of species exhibiting similar characteristics. In taxonomic nomenclature the genus name is used, either alone or followed by a Latin adjective or epithet, to form the name of a spe-

cies.

herbicide A chemical substance used to destroy or inhibit the

growth of plants, especially weeds.

herbivory Feeding on plants.

host The plant or animal on which an organism feeds; the or-

ganism utilized by a parasitoid; a plant or animal suscep-

tible to attack by a pathogen.

host range The different host species that may be utilized by a plant-

or animal-feeding organism.

host specificity The dietary restriction of an organism to a single or

limited food (for herbivores: the number of plant species accepted as food; the highly-evolved, often obligatory association between an insect and its host(s); the degree to which an organism is restricted to a particular number of

plant or animal hosts.)

insect A small arthropod animal that, as an adult, normally has

six legs, three distinct body regions, one pair of antennae,

and one or two pairs of wings.

instar Period or stage between successive molts in a insect larva.

integrated weed management A system for planning and implementing a program to

contain or control an undesirable plant species or group of species, using all available methods and a thorough

knowledge of pest biology.

invasive plant An aggressive and dominant plant, likely to colonize

and become established in new habitats; usually refers to

weeds.

lamina The expanded portion, or blade, of a leaf or petal.

larva (pl. larvae) Immature insect stage between the egg and pupa.

mass rearing The mass production of a natural enemy.

mechanical weed control Mechanical methods that employ physical means to re-

move or control weeds, including activities such as hand pulling, hoeing, tilling, mulching, burning, and mowing.

metamorphosis The change from one life stage to another in insects, such

as from larva to pupa.

molt The process by which insects and other arthropods shed

their exoskeleton ("skin") as they grow and develop; among insects, molting is typically restricted to larval or

nymphal stages.

monoculture An area vegetated by a single plant species.

natural enemies The parasites, predators, pathogens, and other antago-

nists associated with a species of animal or plant that

cause debility or mortality.

no-choice test A test of host-specificity in which the potential biological

control agent is presented with a single, non-target testplant species at a time. Feeding, development, survival rate and/or oviposition rates are recorded and compared

to those on the target weed.

node The position on a stem where leaves or branches origi-

nate; also known as a "joint."

non-target Not being the target of a control method, e.g., not a de-

sired host or food source for a biological control agent.

ocrea (pl. ocreae) Fused stipules that surround the stem at each leaf node;

in mile-a-minute weed the ocreae flare widely into a sau-

cer shape.

oligophagous Feeding on a few (usually related) different types of

plants or prey.

oviposit To lay or deposit eggs.

PABIL Phillip Alampi Beneficial Insect Laboratory, a New

Jersey Department of Agriculature facility for rearing

biocontrol agents, located in West Trenton, NJ.

perennial A plant living more than two years.

petiole A leaf stalk.

physiological host range All plant species that support feeding, development, and

reproduction of a particular insect species, when tested

under laboratory conditions.

phytophagous insect An insect that feeds on plants.

polyphagous insect An insect that feeds on many types of plants or prey.

post-emergent herbicide An herbicide that controls plants via uptake of chemical

through the plant foliage or stems.

pre-emergent herbicide An herbicide that controls plants before they emerge

from the ground by injuring the plant as the seed germi-

nates.

pupa (pl. pupae) (v. pupate) Non-feeding, inactive stage between the larva and adult

in insects with complete metamorphosis.

phylogenetic Relating to or based on evolutionary development or his-

tory.

quadrat A specific area used to sample vegetation (e.g., 1 square

meter, or  $1m^2$ ).

radicle The first part of a seedling (a growing plant embryo)

to emerge from the seed during the process of ger-

mination; the embryonic root of the plant.

scarification Cutting or softening the hard wall of a seed to break seed

dormancy.

seed bank An accumulation in the soil of long-lived seeds, which

can potentially germinate many years after they were

produced.

species A fundamental category of taxonomic classification,

ranking below a genus or subgenus and consisting of

related organisms capable of interbreeding.

surfactant A compound that increases the effectiveness of an herbi-

cide by increasing the adherence of the herbicide mixture to the leaf surface, reducing the surface tension of the mixture so it spreads over more of the leaf and aiding penetration of the waxy outer cuticle of the leaf.

TAG Technical Advisory Group for Biological Control Agents

of Weeds

taxonomy The classification of organisms in an ordered system

that indicates natural relationships. The science, laws, or

principles of classification; systematics.

terminal The growing tip of a mile-a-minute weed vine or vine

branch; may eventually develop into a flower cluster.

thorax Body region of an insect, behind the head, bearing the

legs and wings.

transect A straight line or path through an area.

USDA-APHIS- PPQ United States Department of Agriculture- Animal and

Plant Health Inspection Service- Plant Protection and

Quarantine.

vegetative reproduction Reproduction in plants other than by seeds, such as from

rhizomes, stolons, and from nodes on lateral, often creep-

ing, roots.

viability The proportion of propagules (e.g., seeds) that are alive

and can germinate.

weevil A type of plant-eating beetle, the adults having distinct

snouts of variable lengths.

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