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Monitored releases of *Rhinoncomimus latipes* (Coleoptera: Curculionidae), a biological control agent of mile-a-minute weed (*Persicaria perfoliata*), 2004–2008

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ABSTRACT

Mile-a-minute weed, *Persicaria perfoliata* (L.) H. Gross, is an invasive annual vine of Asian origin that has developed extensive monocultures, especially in disturbed open areas in the Mid-Atlantic region of the United States. A host-specific Asian weevil, *Rhinoncomimus latipes* Korotyaev, was approved for release in North America in 2004, and weevils have been reared at the New Jersey Department of Agriculture Beneficial Insect Laboratory since then. By the end of 2007 more than 53,000 weevils had been reared and released, mostly in New Jersey, but also in Delaware, Maryland, Pennsylvania, and West Virginia. The beetles established at 63 out of 65 sites (96.9%) where they were released between 2004 and 2007, with successful releases consisting of as few as 200 weevils. Weevils were recorded at 30 additional non-release sites in New Jersey, where they had dispersed at an average rate of 4.3 km/year. Standardized monitoring of fixed quadrats was conducted in paired release and control sites at eight locations. Significant differences in mile-a-minute weed populations in the presence and absence of weevils were found at three locations, with reduction in spring densities to 25% or less of what they had been at the start within 2–3 years at release sites, while weed densities at control sites were largely unchanged. Mile-a-minute weed populations at a fourth site were similarly reduced at the release site, but without control data for comparison due to rapid colonization of the paired control site. At the other four locations, all on islands, mile-a-minute weed populations were reduced at both release and control sites without large weevil populations developing, apparently due to environmental conditions such as late frost and extreme drought.

1. Introduction

Mile-a-minute weed, *Persicaria perfoliata* (L.) H. Gross (Polygonaceae; formerly *Polygonum perfoliatum* L.) is an annual vine accidentally introduced from Asia to a nursery near York, Pennsylvania, USA, in the 1930s. It infests natural areas in a variety of habitats in its imported range and has developed extensive monocultures in some areas (Hough-Goldstein et al., 2008a,b; McCormick and Hartwig, 1995; Mountain, 1989; Wu et al., 2002). A biological control program for the weed was initiated by the USDA Forest Service in 1996 (Wu et al., 2002). The most promising candidate agent, *Rhinoncomimus latipes* Korotyaev (Coleoptera: Curculionidae), was tested on plant species in China and in quarantine in Delaware and found to be extremely host specific (Colpetzer et al., 2004a; Ding et al., 2004; Price et al., 2003). This weevil was approved by USDA-APHIS for release in the United States in 2004.

Rhinoncomimus latipes lays its eggs on leaves, stems, and buds of *P. perfoliata*. Larvae feed internally in the stems, and pupate in the soil, producing three to four overlapping generations each year in the Mid-Atlantic region (Lake, 2007). Weevils are active from early spring, as soon as mile-a-minute seedlings appear, until fall, when the vines die with the first hard frost. Simulated damage that mimicked *R. latipes* feeding caused plant mortality when applied to small *P. perfoliata* plants, and reduced biomass and seed production when applied to larger plants (Colpetzer et al., 2004b), suggesting that *R. latipes* could have a substantial impact on *P. perfoliata* populations. Subsequent tests in field cages showed that *R. latipes* feeding on *P. perfoliata* reduces plant growth and delays seed production, and can kill plants when competing vegetation is present (Hough-Goldstein et al., 2008b).

The weevil has been reared at the New Jersey Department of Agriculture Phillip Alampi Beneficial Insect Laboratory in Trenton,

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New Jersey, since fall of 2004. By the end of 2007 more than 53,000 weevils had been reared and released, mostly in New Jersey, but also at sites in Delaware, Maryland, Pennsylvania, and West Virginia. A standardized monitoring protocol was used to track weevil and plant populations at some, but not all, release sites. Here we report on (1) all sites where *R. latipes* were released between 2004 and 2007, noting where the weevil overwintered and established populations; and (2) weevil population growth and impact on the mile-a-minute weed population in selected sites where standardized monitoring has been conducted for at least 3 years.

2. Materials and methods

2.1. Insects and sites

In July 2004, small field releases in Delaware and New Jersey were conducted using *R. latipes* reared in quarantine at the USDA-Agricultural Research Service laboratory in Newark, Delaware, plus additional insects collected and sent from China in 2004 and held in quarantine before release to assure positive identification and absence of pathogens, parasitoids, or other contaminants. All weevils were originally collected from *P. perfoliata* plants at two sites in Hunan Province, China (Leifeng, 28°12′40″ N, 112°50′13″ E and Tongxin, 28°12′21″ N, 112°48′57″ E). Additional insects from the 2004 mixed lab-reared and field-collected group were used to initiate mass rearing at the Phillip Alampi Beneficial Insect Laboratory, where insects were reared on potted *P. perfoliata* plants under fluorescent grow lights (Hough-Goldstein et al., 2008a). All subsequent releases used weevils reared at this facility, except for a few in southeastern Pennsylvania in 2005, which used a combination of weevils reared in a University of Delaware greenhouse and weevils reared at the Phillip Alampi Laboratory, all from the same initial *R. latipes* stock.

For all releases, weevils were brought to the field in 473-ml wax-lined hot beverage cups (Sweetheart Cup Company, Chicago, IL) with holes cut in each end. Nylon mesh was secured over the holes and a plastic Petri dish containing a sponge moistened with honey and water was taped to the bottom of the cups. Excelsior wood shavings (Knud Nielsen Company, Inc., Evergreen, AL) were placed in the cups to give the weevils additional resting sites. For states other than New Jersey, weevils were shipped overnight in these cups packed in styrofoam shipping containers along with a frozen cold pack, and were released as soon as possible following delivery. Upon release, the excelsior and any weevils on it were removed from the cup and placed on mile-a-minute weed foliage, and remaining weevils were either shaken or brushed onto foliage or allowed to walk out on their own.

Weevils were released at 45 sites in New Jersey and 20 sites total in Delaware, Maryland, Pennsylvania, and West Virginia between 2004 and 2007 (Fig. 1), with releases occurring between late April and mid-October each year. In some cases multiple releases were conducted at a single site over the course of the summer. The average size of releases in New Jersey was 1123 ± 169 (mean ± SEM) weevils, with a range of 159–6976 per site, not including one site in the Supawna Meadows National Wildlife Refuge where the New Jersey laboratory had been contracted to release more than 15,000 weevils between June and September 2006. The average size of releases in other states was 793 ± 188 weevils, with a range of 200–3500 total per site. Establishment was assessed as of 2008.

2.2. Monitored sites

Monitoring was conducted at paired release and control sites at two locations in New Jersey (designated NJ-1 and NJ-2), two locations in Delaware (DE-1 and DE-2), one location in Maryland (MD-1), and three locations in West Virginia (WV-1, WV-2, and WV-3), using a standardized monitoring protocol designed to balance ease of implementation with collection of useful plant and insect population data (Blossey and Skinner, 2000). For each location, two sites with similar vegetation and mile-a-minute weed populations, but as far away from each other as possible within the same general area (500 m was suggested), were randomly assigned as either the release or the non-release (control) site. Dates and numbers

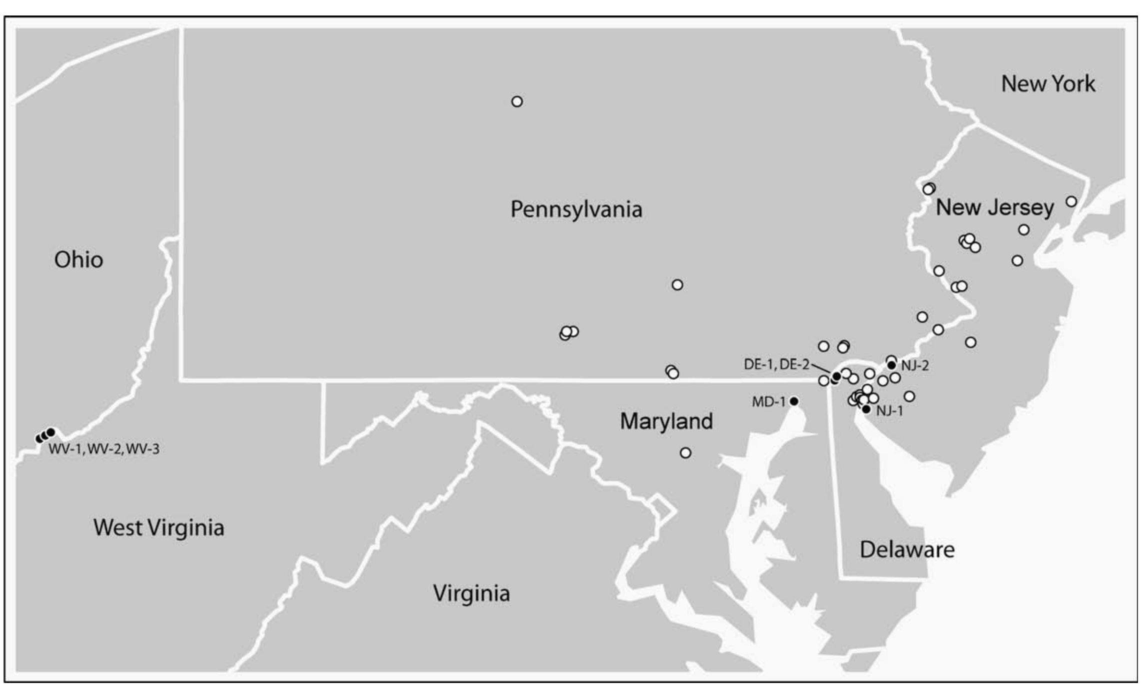

Fig. 1. Sites where *Rhinoncomimus latipes* were released, 2004–2007. Closed circles indicate monitored sites.

of weevils released, and distances between release and control sites for monitored sites are shown in Table 1, and locations are shown in Fig. 1.

At each release and control site, ten 1-m by 0.5-m quadrats, approximately 10 m apart, with ample mile-a-minute weed populations were located and marked at all four corners with pieces of PVC conduit pipe. The quadrats were either located along 100-m-long transects for linear mile-a-minute weed patches (such as along tree lines), or at random within a mile-a-minute weed patch, with quadrat #5, the designated release point for weevil release, located near the center of the patch.

Sites were visited once each year in the spring when mile-a-minute weed seedlings were approximately 10–20 cm tall, generally between late April and mid-May. Numbers of adult *R. latipes*, numbers of mile-a-minute weed seedlings, percent cover of mile-a-minute weed, and presence of *R. latipes* feeding damage was recorded for each quadrat. In 2005, sites were monitored for weevil numbers and damage on 100 mile-a-minute weed terminals (growing tips) surrounding the release site each week following the spring sample. Beginning in 2006, all quadrats were sampled once each month following the spring seedling counts (or first weevil release) until plants senesced. For these samples, sites were surveyed for the following within each quadrat: number of adult weevils; percent defoliation (noting if there were any other insects present aside from *R. latipes* that could have produced the defoliation, such as Japanese beetles); and percent mile-a-minute weed cover (beginning in late summer of 2006 in Delaware and in 2007 at other sites). At each sample date, surveyors were also asked to identify the most abundant other plants in the quadrats.

In New Jersey, the NJ-1 release site was in the Abbott's Meadow Wildlife Management Area and the control site was located 10.6 km away in an isolated overgrown field surrounded by at least a 0.8 km buffer of forest on all sides. All other control sites were located between 250 and 500 m from the paired release site (Table 1). Numbers of weevils released varied from a low of 200 at each of the DE sites (in 2004) and at WV-1 and WV-2 (in 2005) to a high of 6976 at NJ-1 (released between April and September, 2005; Table 1). At site DE-2, in addition to the weevil release, a 1.8 m^3 field cage was placed over mile-a-minute weed plants, with 50 weevils added in August of 2004; weevils reproduced in the cage, which was removed in spring of 2006 when it was apparent that open field releases were very successful. Fewer than 100 additional weevils were released at the DE-1 and DE-2 release sites during the fall of 2005 (left over from nearby cage experiments). The Maryland release site was on Garrett Island, at the mouth of the Susquehanna River. The three West Virginia releases also took place on islands, at the Ohio River Islands National Wildlife Refuge.

For data analyses, the square root transformation (square root $[x + 0.5]$) was applied to count data (weevils and spring seedling counts) and the arcsin transformation (arcsin of the square root of the proportion) to percent cover data, in both cases to reduce heteroskedasticity of variances. Paired release and control sites were compared at each date with *t*-tests; in addition, for the spring samples each release and control site was compared to itself in different years, using ANOVA and LSD comparisons by year. Non-transformed averages and standard errors are shown in Figures.

3. Results

3.1. Weevil establishment and dispersal

As of 2008, adult *R. latipes*, their feeding damage, or both had been observed at 43 out of 45 of the sites where weevils were released in New Jersey in 2004 through 2007, and at all 20 sites where they had been released in other states. In addition to areas where *R. latipes* were released, they were recorded at 30 additional sites in New Jersey where they had dispersed on their own, six between 23 August and 3 September, 2007, and the rest between June and October, 2008. These non-release recoveries in New Jersey occurred between 1 and 3 years following release at the nearest release site (average of 2.4 ± 0.1 years [mean ± SEM]), and averaged 10.7 ± 1.5 km from the nearest release site. Dividing distance by years after release for each non-release recovery site yields an average dispersal distance of 4.3 ± 0.5 km per year.

In southeastern Pennsylvania, weevils were found approximately 200 m from a June 2005 release site in October 2005, and 600–760 m by August 2006 (details in Lake, 2007). In West Virginia, by 2008 *R. latipes* released in 2005 had dispersed from the islands across the Ohio River to both mainlands, and over 8 km up river.

3.2. Monitored sites

All eight of the paired control sites were colonized by dispersing weevils, two within 1 year, and five within 2 years. The NJ-1 control site, which was considerably further away from its paired release site than the others as well as being isolated by a buffer of trees on all sides, remained apparently weevil-free for 3 years (Table 1). Assuming the colonizers at each control site came from its paired release site, the weevils that moved into these sites had dispersed at a rate of 125–500 m per year for most sites, and at 3.5 km per year for the NJ-1 site (Table 1).

At the NJ-1 site, spring mile-a-minute weed densities at the release and control site were comparable at the start of the study, but by 2007 weed densities at the release site were only about 25% of what they had been at the start, while densities at the control site were largely unchanged (Fig. 2). Spring seedling counts and percent cover were significantly lower in the release than in the control plots in 2006, 2007, and 2008 (Fig. 2). Following release

Table 1
Location, date and numbers of weevils released at monitored release sites, distance to paired control sites, and date and time until weevils were first found in control plots.

Site	Location	Dates released	Number released	Distance to control	First weevils in control	Time to reach control (yr)	Dispersal rate per year
NJ-1	Abbott's Meadow WMA	4/22–9/9/05	6976	10.6 km	7/22/08	3	3.5 km
NJ-2	Floodgate Road	7/28/04	200	500 m	9/27/06	2	250 m
		6/17–8/29/05	3297				
DE-1	White Clay Creek State Park	7/21, 7/28/04	200	250 m	6/5/06	2	125 m
DE-2	White Clay Creek State Park	7/21, 7/28/04	200	300 m	6/5/06	2	150 m
MD-1	Garrett Island	6/16/06	500	300 m	8/15/07	1	150 m
		6/18/07	600				
WV-1	Muskingum Island	6/10/05	200	500 m	7/10/07	2	250 m
WV-2	Muskingum Island	6/10/05	200	500 m	7/10/07	2	250 m
WV-3	Neal Island	6/16/06	1000	500 m	7/6/07	1	500 m

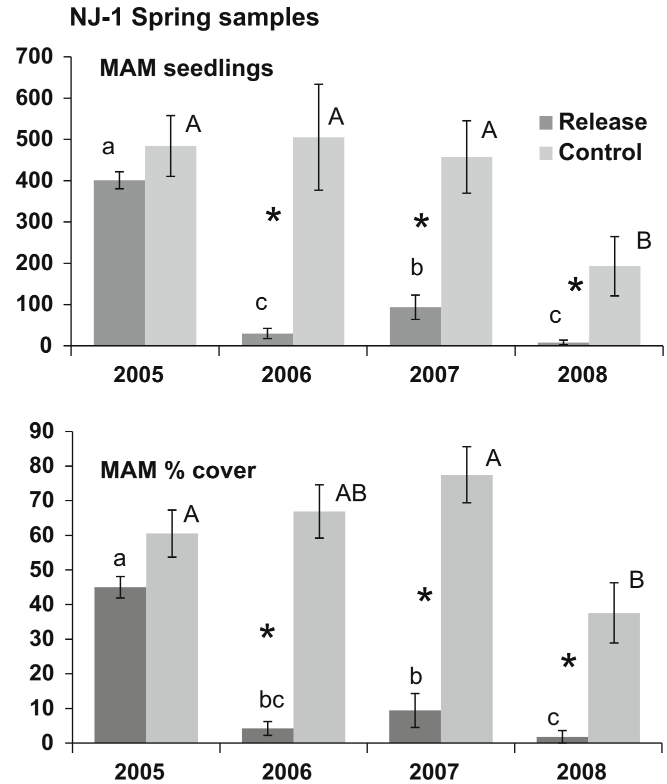

Fig. 2. Mile-a-minute weed seedlings and percent cover in May, site NJ-1. Bars with the same letter (lower case for release, upper-case for control) are not significantly different from each other over different years; paired release and control bars each year with an asterisk are significantly different from each other ($P \leqslant 0.05$, ANOVA and LSD comparisons conducted on square root-transformed seedling counts and arcsin-transformed percent cover data; original means and SEMs shown).

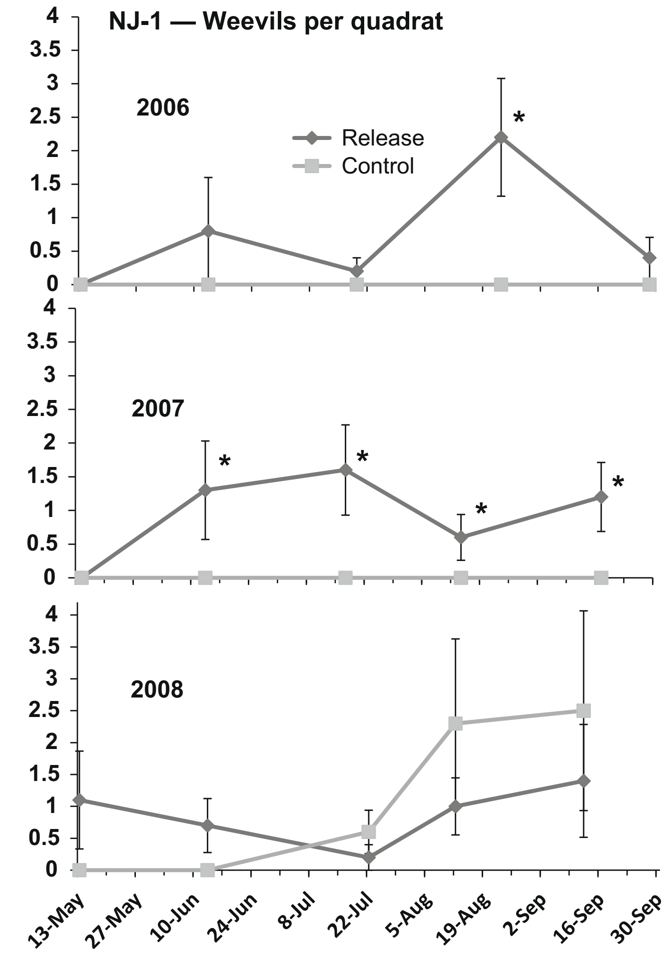

Fig. 3. *Rhinoncomimus latipes* adults per quadrat, site NJ-1. Asterisk indicates significant difference between release and control quadrats ($P \leqslant 0.05$, *t*-tests conducted on square root-transformed weevil counts; original means and SEMs shown).

of nearly 7000 weevils during the summer of 2005, a substantial weevil population developed at the NJ-1 release site by August, 2006 (Fig. 3), and heavy feeding by these weevils depleted the available mile-a-minute weed foliage and appeared to trigger weevil dispersal. Weevil numbers remained significantly higher in the release than in the control site throughout 2007, but by 2008 weevil counts did not differ between control and release sites (Fig. 3). Percent cover of mile-a-minute weed was significantly lower in the release than in the control site throughout 2007 (2 years post-release) and until August 2008 (Fig. 4).

The NJ-2 site received the second largest number of weevils after NJ-1, with a total of 3497 released over 2 years, 400 in 2004 and the rest in 2005 (Table 1). Weevils were found at the NJ-2 control site by August 2006, and monitoring was subsequently discontinued at this site. At the release site, the spring mile-a-minute weed population declined from more than 290 seedlings (56% cover) per quadrat in 2005 to fewer than 32 (12% cover) in 2008 (Fig. 5). Weevil numbers were very high at this site, with an average of more than 11 per 0.5-m^2 quadrat counted in August of 2006 (Fig. 6), causing obvious defoliation of the mile-a-minute weed. By October, 2006, plants were severely damaged and numerous weevils were present: more than 200 weevils were collected in under a minute by placing a clipboard under the defoliated stems at this site and gently tapping the plants. Weevil numbers declined in monitored quadrats during 2007 and 2008 (Fig. 6), as the percent cover of *P. perfoliata* in the NJ-2 release quadrats declined to 15% or less (Fig. 7).

At the DE-1 release site, as at NJ-1, mile-a-minute weed population densities were comparable in the release and control sites in spring of 2005, but by 2006 the release site quadrats averaged less than 5% cover, while seedling numbers and percent cover remained unchanged at the control site (Fig. 8). Both seedling counts and spring percent cover were significantly lower in the release than in the control quadrats in 2006 and 2007 (Fig. 8). By 2008 very little mile-a-minute could be found at the DE-1 release site except at the sunny, exposed edges of the patch bordering a mowed field. Summer monitoring at this site was discontinued in 2007 and 2008 due to a lack of mile-a-minute weed in the monitored quadrats. Observations in 2008 suggested that the few mile-a-minute weed plants that continued to emerge in the center of this diverse site were heavily defoliated by *R. latipes*. Numerous weevils and considerable feeding damage was also observed on the mile-a-minute weed plants present at the edge of the site. By 2007 weevils were present at relatively high numbers in the DE-1 control plots (Fig. 9). The DE control plots were only sampled once during the summer of 2008, on 13 August. At that time the mean (±SEM) percent cover was 15.4 ± 8.5% at the DE-1 control site (now with many weevils present), significantly lower than percent cover in these quadrats the previous year (14 August, 2007, 50.5 ± 6.7%; $F_{1,12} = 22.23$, $P = 0.0005$).

At the DE-2 release site, where 200 weevils were released in late July 2004, mile-a-minute weed seedling numbers were significantly higher at the release site than at the control site in May of 2005 (the first sample), suggesting that the two sites did not have equivalent mile-a-minute weed populations at the start of the experiment. However, over the next 3 years there was a gradual

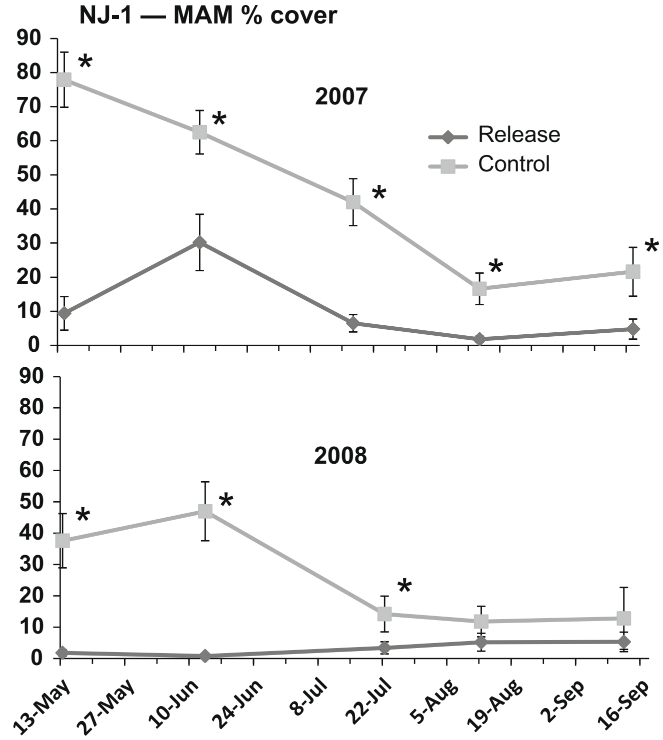

Fig. 4. Mile-a-minute weed percent cover, site NJ-1. Asterisk indicates significant difference between release and control quadrats ($P \leqslant 0.05$, *t*-tests conducted on arcsin-transformed percent cover data; original means and SEMs shown).

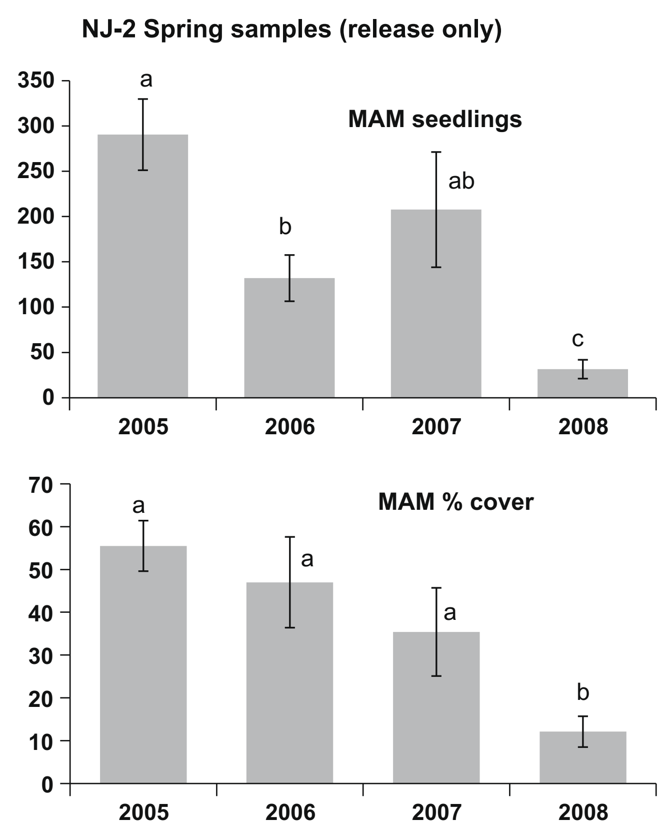

Fig. 5. Mile-a-minute weed seedlings and percent cover in May, site NJ-2, release site only. Bars with the same letter are not significantly different from each other over different years ($P > 0.05$, ANOVA and LSD comparisons conducted on square root-transformed seedling counts and arcsin-transformed percent cover data; original means and SEMs shown).

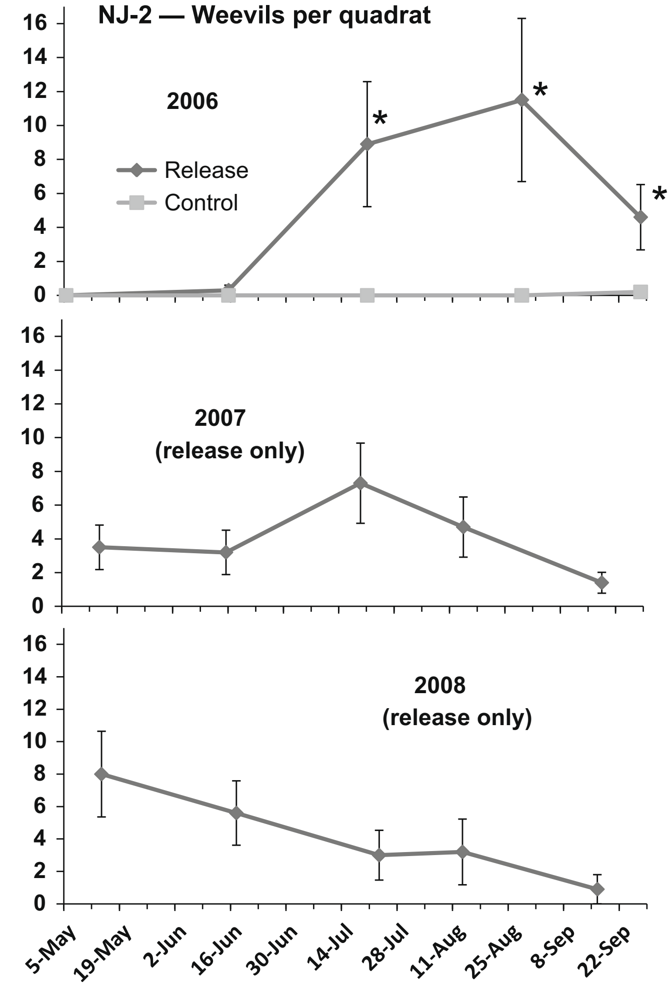

Fig. 6. *Rhinoncomimus latipes* adults per quadrat, site NJ-2. Asterisk indicates significant difference between release and control quadrats ($P \leqslant 0.05$, *t*-tests conducted on square root-transformed weevil counts; original means and SEMs shown).

reduction in the numbers of *P. perfoliata* seedlings and percent cover in the spring samples at the release site, from over 200 per quadrat (54% cover) in 2005, to fewer than 14 (12% cover) in 2008 (Fig. 10), while seedling numbers in the control site did not differ significantly between 2005 and 2007. Spring percent cover was lower at control site DE-2 in 2006 than in 2005, but subsequently increased at this site in 2007 (Fig. 10). Weevils were abundant at the release site in 2006 through 2007 and were present but at generally lower numbers each year at the control site (Fig. 11). Weevil counts in August 2008 averaged 9.6 ± 3.6 per quadrat at the release site and 0.7 ± 0.5 at the control site. Percent cover of mile-a-minute weed remained relatively high at both sites through 2007 (Fig. 12), and in mid-August of 2008 was still 32.1 ± 9.3% at the release site, not significantly different from the control (17.8 ± 8.0%) or from the release site the previous year (14 August, 2007: 42.5 ± 5.6%; $F_{1,15} = 1.21$, $P = 0.2885$).

At MD-1 on Garrett Island, the mile-a-minute weed population was less dense than at the New Jersey and Delaware sites, with spring seedling counts averaging 69 per quadrat in 2006. Although weevils were found near the release site in 2006 during the summer following the spring release and in the monitored quadrats in 2007 and 2008, numbers remained low (data not shown). Conditions were extremely dry during the summer of 2007, causing mile-a-minute weed plants to die back at both the release and the control plots at this site, with percent cover averaging only

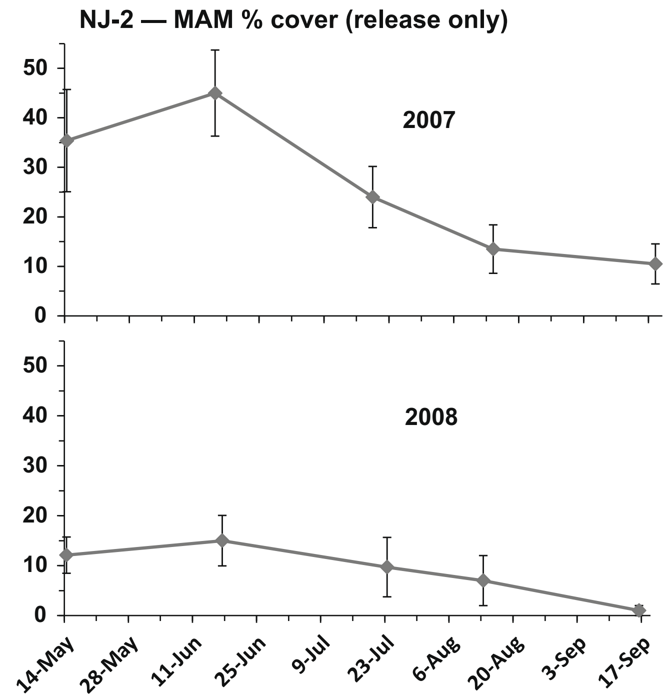

Fig. 7. Mile-a-minute weed percent cover, NJ-2 release site.

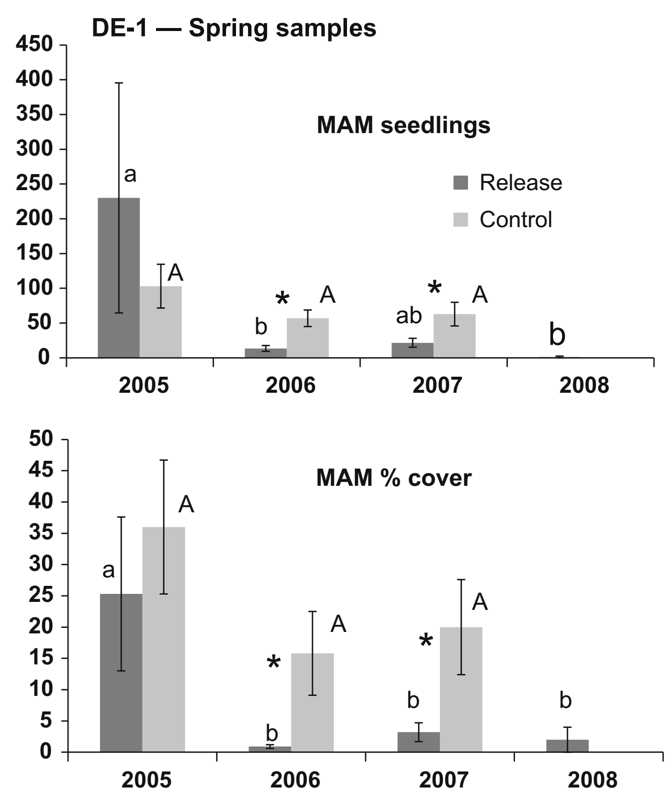

Fig. 8. Mile-a-minute weed seedlings and percent cover in May, site DE-1. Bars with the same letter (lower case for release, upper-case for control) are not significantly different from each other over different years; paired release and control bars each year with an asterisk are significantly different from each other ($P \leqslant 0.05$, ANOVA and LSD comparisons conducted on square root-transformed seedling counts and arcsin-transformed percent cover data; original means and SEMs shown).

4–5% at all sites by July 2007. Weevils were found at the control site within 1 year of release.

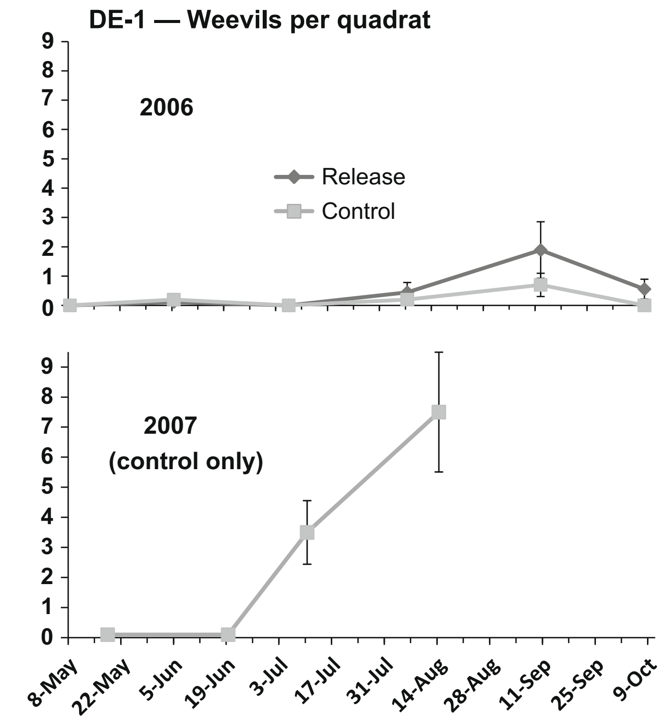

Fig. 9. *Rhinoncomimus latipes* adults per quadrat, site DE-1. Asterisk indicates significant difference between release and control quadrats ($P \leqslant 0.05$, *t*-tests conducted on square root-transformed weevil counts; original means and SEMs shown).

Similar results were found at the Ohio River Island NWR sites in West Virginia. Although seedling numbers and percent cover were fairly high in May of 2005 and 2006, in the spring of 2007 a late frost apparently killed many plants in all release and control plots (data not shown). By August of 2007, the percent mile-a-minute weed cover averaged between 1% and 18% at all WV sites. Weevil numbers were also low, and were found at all control sites within one or 2 years (Table 1).

4. Discussion

Rhinoncomimus latipes weevils were able to overwinter and establish field populations in 63 out of 65 sites (96.9%) where they were released between 2004 and 2007, ranging from northern New Jersey west to the Ohio River Valley and south to Howard County, Maryland. Their hardiness and ability to thrive in a variety of settings was predicted, because in China they are widely distributed in nine provinces from north to south, including Heilongjiang province in northeastern China, where winter temperatures can reach lows of minus 30–40 °C (Ding et al., 2004). The weevil populations were also able to withstand extremes of flooding. One of the 2005 New Jersey release sites was located near the Delaware River and experienced a "100-year flood" in the spring of 2006. The high waterline was 0.5 m above the release site, and weevils were not expected to survive, but *R. latipes* adults were observed at the release site in late May that year and in subsequent years. Weevils also persisted at another 2005 release site along the Delaware River in Salem County, New Jersey, which was periodically flooded by tides. In its native range in Asia, *P. perfoliata* is primarily a riparian species and thus may be adapted to survive annual flooding (Hyatt and Araki, 2006).

The two releases where weevils did not successfully overwinter both took place in October (one in 2006 and one in 2007), suggesting that weevils released this late in the year may not have suffi-

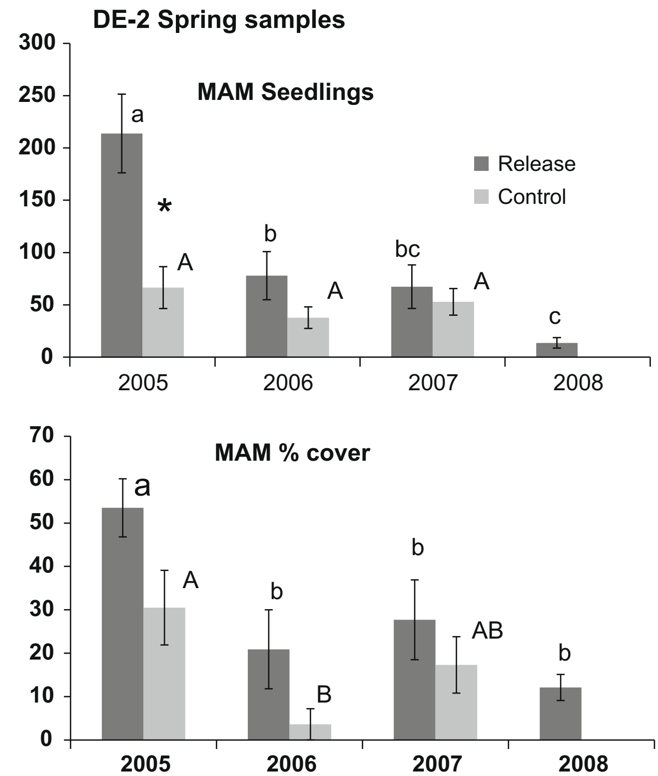

Fig. 10. Mile-a-minute weed seedlings and percent cover in May, site DE-2. Bars with the same letter (lower-case for release, upper-case for control) are not significantly different from each other over different years; paired release and control bars each year with an asterisk are significantly different from each other ($P \leqslant 0.05$, ANOVA and LSD comparisons conducted on square root-transformed seedling counts and arcsin-transformed percent cover data; original means and SEMs shown).

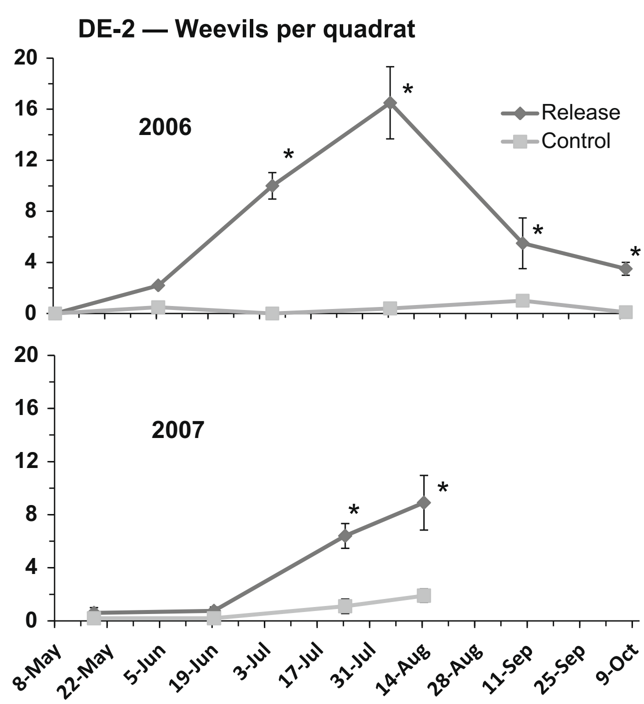

Fig. 11. *Rhinoncomimus latipes* adults per quadrat, site DE-2. Asterisk indicates significant difference between release and control quadrats ($P \leqslant 0.05$, *t*-tests conducted on square root-transformed weevil counts; original means and SEMs shown).

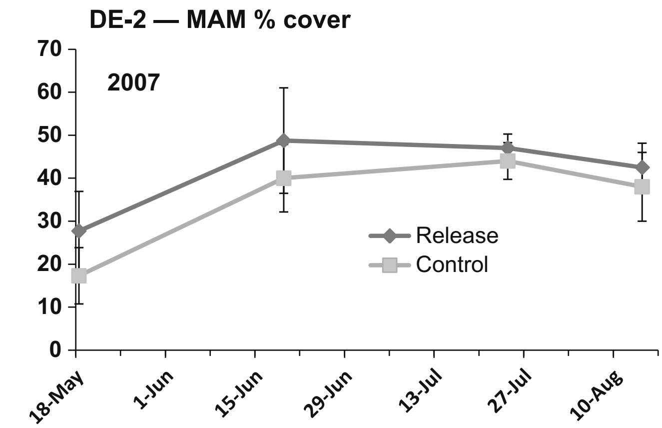

Fig. 12. Mile-a-minute weed percent cover, site DE-2 (release and control quadrats did not differ significantly; $P > 0.05$, *t*-tests conducted on arcsin-transformed percent cover data; original means and SEMs shown).

cient time to acclimate before winter's onset. *R. latipes* stops laying eggs between mid-August and late September in southeastern Pennsylvania (Lake, 2007), but adults continue to feed on mile-a-minute weed foliage in preparation for overwintering. Two other releases in 2006 that were conducted on the same date in October (and with the same number of weevils, 600) as the one that failed, however, did establish, and thus lack of time for acclimation was not the sole reason for failure. Conditions were somewhat rocky and dry at the site that failed, which may have contributed to weevil overwintering mortality. The 2007 release that failed was conducted with older laboratory weevils, probably less vigorous than the newly emerged weevils typically used for releases, which may have contributed to this failure (as found, for example, by Center et al. (2006), for a psyllid biological control agent). This release was also the smallest, with only 159 individuals released, and thus low propagule size may have contributed to failure (e.g., Memmott et al., 2005).

At all release sites where monitoring occurred, paired control sites were also established, in an attempt to measure the effects of weevil feeding on mile-a-minute weed populations compared to similar populations without the weevil, as recommended by McClay (1995) and more recently by Carson et al. (2008), among others. Seven of the eight control plots were colonized within 2 years, with an average dispersal rate of 125–500 m/year. The most remote control plot, however, was found in 3 years, indicating dispersal at an average rate of 3.5 km/year, consistent with the average dispersal rate of 4.3 km/year estimated from 30 non-release recovery sites in New Jersey, most of which were discovered 2 or 3 years after the nearest releases had been made. The 4.3 km/year estimate is conservative, since the weevils may have arrived sooner than when they were discovered. These observations were not part of a systematic effort to track dispersal, but generally occurred when staff from the New Jersey Beneficial Insect Laboratory visited a new mile-a-minute site in preparation for releasing weevils and found that it was already colonized.

This rate of dispersal is greater than that of several other biocontrol agents. For example, the weevil *Oxyops vitiosa* (Pascoe), introduced for control of *Melaleuca quinquenervia* (Cav.) Blake in South Florida, dispersed at a rate of 0.1–2.8 km/year (Pratt et al., 2003). *Aphthona nigriscutis* Foudras (Coleoptera: Chrysomelidae) released for control of leafy spurge in Canada spread at a rate of only 5–10 m per year in the first few years (McClay, 1995). The spread of *R. latipes* was similar to that of the psyllid *Boreioglycaspis melaleucae* Moore, which spread at an average rate 4.7 km/year (Center et al., 2006). Despite relatively rapid dispersal, redistribution efforts, especially to mile-a-minute weed patches at the edges of the plant's current distribution, in Rhode Island, Connecticut, Massachusetts, and New York State to the north and Virginia to the south, would be beneficial and in some cases are already underway.

Despite dispersal of the weevils into the controls, significant differences in mile-a-minute weed populations in the presence and absence of the weevil were found at three sites, NJ-1, DE-1, and DE-2. At both NJ-1 and DE-1, mile-a-minute weed densities at the release and control sites were comparable at the start of the study in 2005, but by the end of 2007 weed densities at the release site were 25% or less of what they had been at the start of the study, while densities at the control site were largely unchanged. At site DE-2, the control site had a lower mile-a-minute weed population than the release site initially, but did not change in seedling numbers for 3 years, while seedling numbers at the release site were reduced to about 25% of their initial density. The less convincing but more conventional before-and-after comparisons at site NJ-2 (where the control site was abandoned following rapid weevil colonization) also showed significant reductions in the mile-a-minute weed population along with large increases in weevil numbers over 3 years.

The design used here was similar to the "Before-After-Control-Impact" (BACI) design described by Osenberg and Schmitt (1996) for assessing ecological impacts of human-caused perturbations, in this case release of weevils. With this design, spatial variability among field sites is controlled by looking at the same site over time (before and after), and temporal variability, i.e., changes that will occur at all sites over time whether the impact of the human perturbation is present or not, is controlled for by monitoring control sites over the same time period as the impacted sites. This design proved valuable here despite fairly rapid weevil dispersal into the controls, and provided convincing evidence of the impact of *R. latipes* on mile-a-minute weed populations over 2–3 years at several different locations.

At both the Garrett Island, Maryland, and the Ohio River Islands, West Virginia, sites, the mile-a-minute weed populations collapsed within all monitored quadrats, release and control sites, after 1 or 2 years. The reduced mile-a-minute weed populations were probably due largely to environmental factors such as severe drought and late spring frost at these sites, rather than to effects of weevil feeding. Hyatt and Araki (2006) similarly found that in permanent quadrats starting with relatively low densities of mile-a-minute weed seedlings (and before any insects had been released) plant population growth rates were highly variable, with several populations showing negative growth rates within the quadrats. It may be that mile-a-minute weed is a relatively poor competitor when it is present initially at low densities and in the presence of competing plant species, and depends on disturbance and dispersal of its large seed crop to maintain a metapopulation.

Results from monitored sites suggest that, once established, *R. latipes* is capable of producing the population outbreaks that are often associated with successful biological control of weeds (Van Driesche et al., 2008). The monitoring protocol likely produced an undercount of actual weevil numbers, because adults are very small (~2 mm long), sometimes rest or feed on the underside of leaves or hidden in terminals, and may drop to the ground when disturbed. Nevertheless, enough weevils were visible to allow for counts of up to 11 (at NJ-2) to 16.5 (at DE-2) on average per 0.5-m^2 quadrat. In this study weevil counts were not adjusted based on the percent cover of mile-a-minute weed in the quadrats, as was done, for example, by Lake (2007) and thus as cover declined, weevil counts per quadrat also tended to decline.

The monitoring protocol based on permanent quadrats, as recommended by Blossey and Skinner (2000), proved feasible for cooperators at various sites and provided an adequate assessment of changes in the plant and insect communities over time. Comparisons between control and release sites, however, will become less feasible as *R. latipes* continues to disperse throughout the range of its host plant.

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