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DISPERSAL AND DISTRIBUTION OF BIOLOGICAL CONTROL AGENTS FOR *LYTHRUM SALICARIA* IN INDIANA

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ABSTRACT. The invasive wetland perennial, *Lythrum salicaria* has spread throughout Indiana wetlands since 1900. Four insect species were approved for release as biological control agents. These species included: *Hylobius transversovittatus*, *Nanophyes marmoratus*, *Galerucella calmariensis*, and *G. pusilla*. The distribution of these beetles has been monitored by the Indiana Department of Natural Resources since 1994. This project aimed to develop an updatable GIS database, expand the existing data on locations of these control agents, and estimate their distribution throughout the state. *Nanophyes marmoratus* and *Hylobius transversovittatus* have spread slowly between wetlands, which limited the analysis of this work to the spread of *Galerucella*. Geospatial analyses of *Galerucella* spp. indicates that they have become widely distributed in the northern region of the state. By calculating distances and date of initial observation between sites, it was estimated that *Galerucella* spp. spread at a rate of least 491 meters per year with a maximum rate of 1,822 m/yr. This simplified calculation of dispersal rates and GIS mapping allows for visualization of areas for potential future releases in order to maximize the control of *L. salicaria*. Additionally, it suggests that *Galerucella* spp. have become widely established in northern Indiana.

Keywords: *Lythrum salicaria*, *Galerucella*, *Nanophyes marmoratus*, *Hylobius transversovittatus*, biological control, invasive species, geospatial distribution

Lythrum salicaria, or purple loosestrife, is an invasive wetland perennial in North America (Thompson et al. 1987). It can form clumps of 30–50 stems arising from a single taproot and a terminal spike of tightly clustered flowers over 1 meter in length (Mal et al. 1992). This vigorous branching and flowering allows a single plant to produce upwards of 2.5 million seeds each year (Malecki et al. 1993).

North American introductions of *L. salicaria*, which began as early as 1814 in New England (Mal et al. 1992), were likely from inadvertent transport in shipping ballast and imported wool and intentional introduction by immigrants, who used the plant as a medicinal herb (Thompson et al. 1987). Anthropogenic activities, such as the development and use of canals, contributed to the further spread of *L. salicaria* into the Midwest (Thompson et al. 1987). Other uses of the plant, both as an ornamental and as a nectar plant for beekeeping, resulted in intentional introductions. In Indiana the earliest record of *L. salicaria* is from 1900, although Stuckey (1980) and Deam (1940) note very little spread through 1940. Today, *L. salicaria* is distributed throughout Indiana but is most common in the northern counties.

Lythrum salicaria forms dense stands (Malecki et al. 1993) that can become dominant in wetland seed banks (Welling and Becker 1990), reduce wildlife habitat quality (Whitt et al.1999, Rawinski 1982, Lor 2000), interfere with pollination of native species (Brown et al. 2002, Da Silva and Sargent 2011, Templer et al. 1996), and alter wetland function (Emery and Perry 1996, Bärlocher and Biddiscombe 1996). It may also outcompete native plants in a variety of wetlands (Gaudet and Keddy 1995, Weihe and Neely 1997, Gabor et al. 1996, Mal et al. 1997).

Control efforts against *L. salicaria* began in the 1950's, and included attempts at flooding, cutting, and burning (Skinner et al. 1994). Initial efforts were largely unsuccessful for all

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but the smallest patches of *L. salicaria* (Blossey et al. 2001, Skinner et al. 1994). Glyphosate, 2,4-D, or triclopyr have been used in chemical control but because of extensive seed banks (Welling and Becker 1990), spraying must be repeated often (Blossey et al. 2001, Skinner et al. 1994). The non-selective nature of chemical control also reduced populations of sedges, grasses, cattails, and other native wetland plants (Skinner et al. 1994, Gabor et al. 1996). The lack of effective methods of control and continued dispersal of *L. salicaria* resulted in the formation of a program to establish a biological control program for *L. salicaria* in North America. An overview of this process is provided by Malecki et al. (1993), while Blossey et al. (2001), and Hight and Drea (1991) examine the process in more detail. Four insect species eventually were approved for release. These species included: *Hylobius transversovittatus* Goeze (a root-mining weevil), *Nanophyes marmoratus* Goeze (a flower feeding weevil), *Galerucella calmariensis* L., and *G. pusilla* Duft (two leaf-beetles) (Blossey et al. 2001).

Following the approval of *Galerucella* spp., *N. marmoratus*, and *H. transversovittatus* for release, all three were released at sites in Indiana by the Indiana Department of Natural Resources (IDNR), Division of Nature Preserves. *Galerucella* spp., the earliest to be approved, were first released in 1994. Release of *N. marmoratus* and *H. transversovittatus* did not begin until eight years later, in 2002. A protocol for raising captive populations of *Galerucella* was quickly established. As releases of *Galerucella* spp. continued, and as populations accumulated, individuals were collected from existing populations and released to sites where they were previously absent. Thus, of the three genera of insects, *Galerucella* spp. were most widely released in Indiana because they were approved first and were easily raised.

The aim of this project was to develop an easily updateable geodatabase for tracking the distribution of purple loosestrife biological control agents across the state. The development of this database is important for improving our understanding of the specific dispersal range of *Galerucella* spp. as well as the general dispersal patterns of biological control agents. It was also a goal of this project to understand the current spread and patterns of dispersal, especially of *Galerucella* spp., by field checking numerous sites throughout northern Indiana in 2011 supplemented by 17 years of continued observations made by IDNR personnel. Furthermore, knowledge of the current range of *Galerucella* spp. is important for the planning of future releases of the insects in order to maximize the effectiveness of the biological control program for *L. salicaria*.

METHODS

The primary files in the geodatabase are point feature classes of locations where beetle populations have been located. To maintain these records, any new release site is added to the files and time each spring is spent checking locations where the beetles are likely to have spread. This surveying involves driving to areas around known release or dispersal sites and identifying areas infested by *L. salicaria*. Once *L. salicaria* is found, the area is checked for presence of beetles and their abundance is ranked.

During the summer of 2011, particular effort was made to survey a number of sites within spatial gaps in the current data. In order to check a large number of sites over a short period of time most of the sites examined were boat ramps. In addition to ease of checking for *L. salicaria* and biological control agents, boat ramps are disturbed habitats, often are in full sunlight (favored by *L. salicaria*), and boats and trailers provide a means of spreading loosestrife seed. As a result of this additional work, 103 sites were examined, with nearly all of them being boat ramps.

Geospatial analysis, processing, and map construction was performed using ArcGIS Desktop 10 (ESRI 2011). In addition, the Geospatial Monitoring Environment (Beyer 2012) was used to calculate distances between release and non-release sites. A point feature class file containing the sites where insects were located was created for each beetle species. These files also contained the tabular data for each location, including whether insect were released, the county, insect abundance, first observation date, and latest observation date. A series of maps were created to show the dispersal of *L. salicaria* biological control insects in Indiana.

Finally, the geospatial data were analyzed to determine the likely dispersal rate of *Galerucella* spp. Non-release sites were joined to each release site manually, using data exported to spreadsheets. Using the *convert.tabletolines*

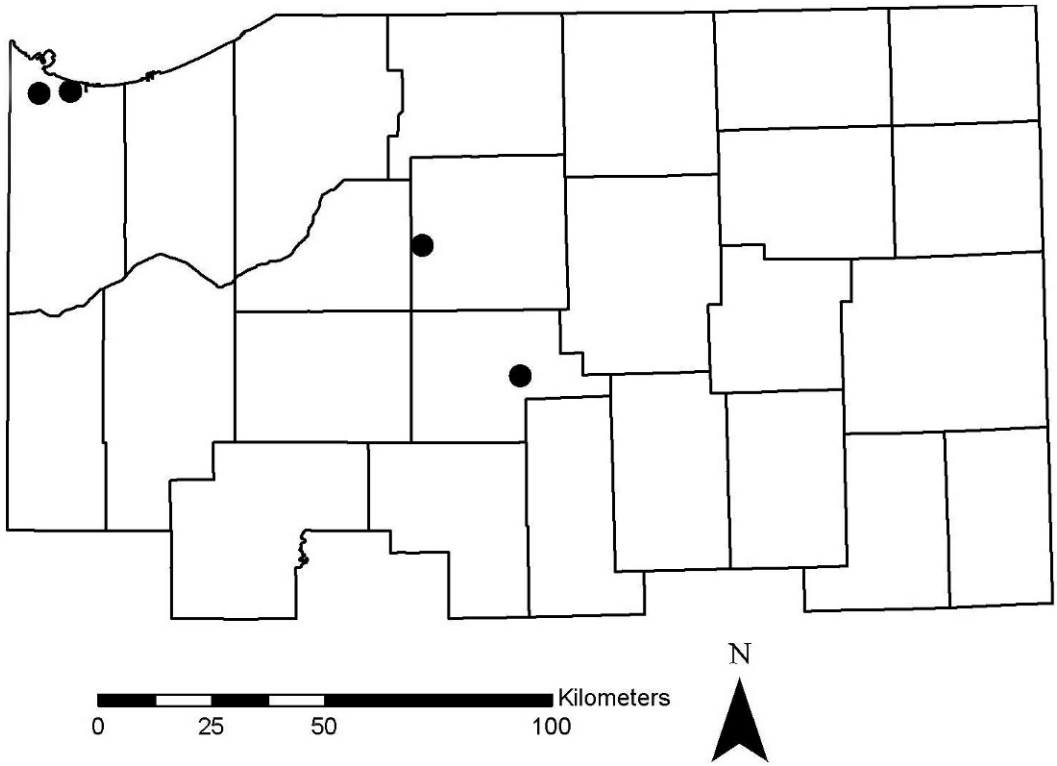

Figure 1.—*Hylobius transversovittatus* sites in Indiana. All sites are release sites.

command in the Geospatial Modeling Environment (Beyer 2012), the distance between each set of points was determined and added to the table. For each pair of sites the number of years between the release of *Galerucella* spp. at the release site and the first observation at the non-release site was determined. The distance between sites was divided by the years between these dates to calculate the average distance traveled per year. The assumption was made that the smallest distance travel per year was the most likely source of the beetles at the new site, and other pairs of points were removed. One record remained for each non-release site. The maximum distance per year (1,822 m/yr) and the mean distance per year (491 m/yr) from this list were then used to draw buffers around known *Galerucella* spp. locations in ArcMap. The calculated dispersal distance per year was multiplied by the number of years since the first observation (or release date) at each site. The buffer was then drawn at this distance. The buffer distance around a newer site is smaller, while that of an old site is much larger.

RESULTS AND DISCUSSION

This project sought to examine the distribution of all three biological control insects throughout Indiana. However, locating *H. transversovittatus* proved to be very difficult. They have not been located at new sites (Figure 1) and only occasionally at past release sites; this is likely due to their secretive life cycle (Ferrarese and Garono 2010). The adult beetles are nocturnal (Blossey et al. 1994), so surveying is usually conducted for beetles in the larval stage. Since this stage is completed below ground, surveying is labor intensive and requires uprooting the plants for close examination (Malecki et al. 1993). Therefore, it is difficult to draw conclusions as to their distribution within Indiana.

Nanophyes marmoratus have been found at some non-release sites, but all of these were located near a release site, suggesting that their dispersal has been limited (Figure 2). In addition, their dispersal may be limited by the success of *Galerucella* spp. Large populations of *Galerucella* spp. may reduce the flowering of

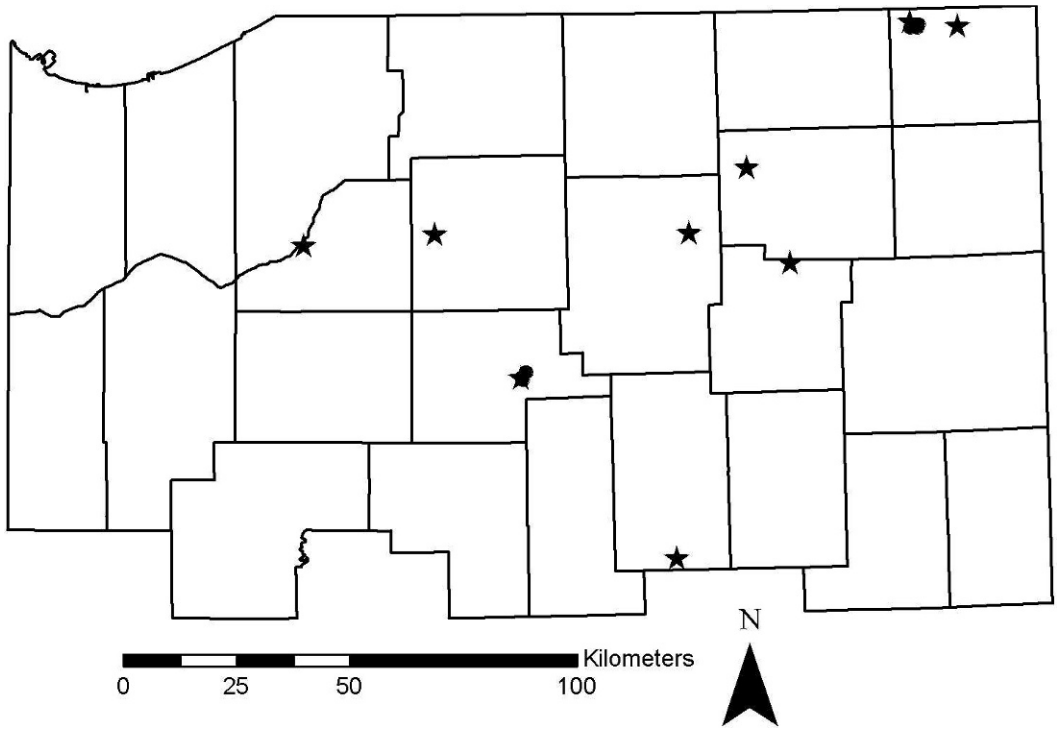

Figure 2.—*Nanophyes marmoratus* release sites (★) and non-release (●) in Indiana.

L. salicaria (Blossey and Skinner 2000), flowers necessary to the life cycle of *N. marmoratus*. Additionally, *N. marmoratus* has been release at far fewer sites in Indiana than *Galerucella* spp., further reducing the potential for locating new sites for *N. marmoratus*.

In contrast to *Hylobius* and *Nanophyes, Galerucella* spp. have dispersed quite well and were relativity easy to locate, resulting in 156 confirmed sites (Figures 3). In particular, *Galerucella* spp. have spread across northern Indiana but, in the remainder of the state where *L. salicaria* is infrequent, has only 5 reports (one in Brown, Marion, and Scott Counties; two in Morgan County). The mean annual dispersal distance of *Galerucella* spp. was calculated at 491 m/yr and the maximum was 1,822 m/yr. The buffered areas in Figure 4 approximate the potential area to which *Galerucella* spp. may have dispersed. These areas cover a total of 5,294 km^2 for the dispersal of 491 m/yr and 33,348 km^2 based on 1822 m/yr. Using the National Wetland Inventory as a basis for *L. salicaria* habitat, the potential *Galerucella* spp. habitat is reduced greatly to 747 km^2 (491 m/yr) and 2,799 km^2 (1822m/yr).

When compared to previous studies, these dispersal estimates fall within the wide range of values observed. Albright et al. (2004) found *Galerucella* spp. 9 km from the nearest release site after 4 years. If the dispersal is estimated over four years, the distance of 2,250 m/yr is slightly larger than the 1822 m/yr found in this study. However, Dech and Nosko (2002) suggest a very limited dispersal rate of both *Galerucella* species. After four years *Galerucella* spp. were found a maximum of approximately 50 meters from the release location.

Based upon additional observations made during the surveying for *Galerucella* spp. it appears that the beetles are well distributed to potential habitat within the 491 m/yr buffer area. Dispersal throughout the larger buffer area is more sporadic, but a large number of sites do occur (Figure 4). While the assumption was made that *Galerucella* spp. dispersal occurs in a uniform fashion over time (x m/yr) this is almost certainly not the case. This assumption allows for an approximation of the potential

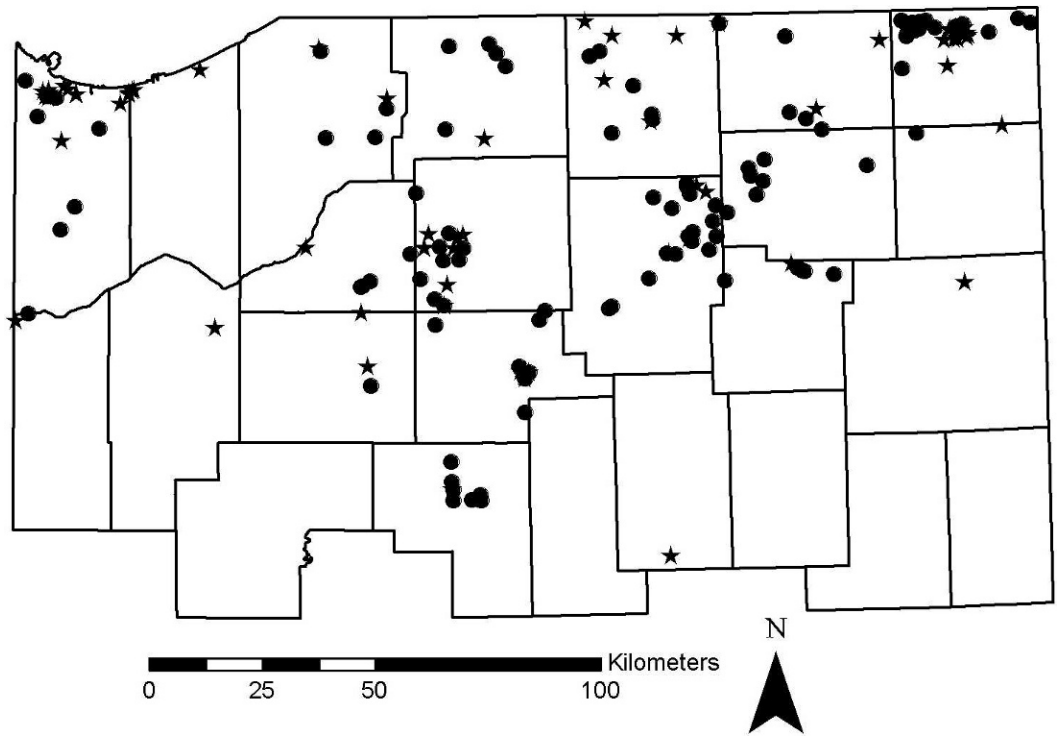

Figure 3.—*Galerucella* spp. release sites (★) and non-release sites (●) in northern Indiana.

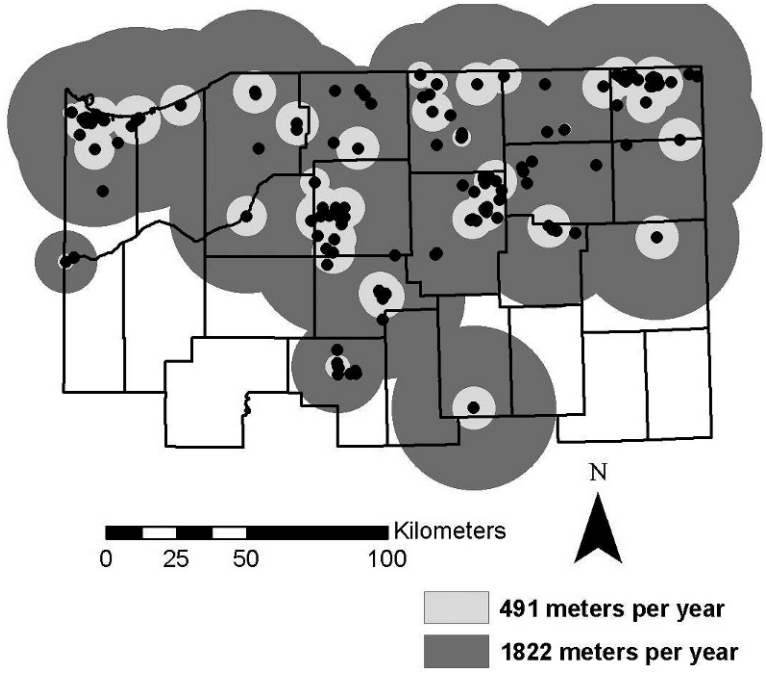

Figure 4.—*Galerucella* spp. release sites and non-release sites in Indiana with buffers showing potential area currently occupied by *Galerucella* spp. The small buffer area is based on 491 m/yr and the larger on 1822 m/yr.

area inhabited by *Galerucella* spp., but their dispersal patterns are more complex. Bartelt et al. (2008), Hambäck (2010), and Grevstad and Herzig (1997) have shown the importance of pheromones released by *Galerucella* spp. and the resulting aggregation behavior. Additionally, when large *Galerucella* spp. populations accumulate they can defoliate nearly all *L. salicaria* in the area (Landis 2003). Following this defoliation, beetles have been observed to disperse in large numbers to new sites, further complicating the dynamics and patterns of dispersal.

While the areas defined in Figure 4 are potential areas of occurrence, they provide an approximation for future biocontrol action. Using these maps, the areas least likely to be currently occupied by *Galerucella* spp. can be identified and additional releases can be performed in these areas. Additionally, the results of this study improve our understanding of the dispersal range of *Galerucella* spp. over time. Future work will be able to continue adding new locations to the geodatabase and as data are added the estimations of dispersal range will improve. Additionally, because the methodologies are easily applied to any case where the release locations have been documented, the methods used here may be applied in other locations where *Galerucella* spp. have been released in order to understand any regional differences that may occur.

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