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A field-based method to estimate densities of *Laricobius nigrinus* (Coleoptera: Derodontidae), an introduced predator of hemlock woolly adelgid (Hemiptera: Adelgidae)

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The hemlock woolly adelgid, *Adelges tsugae* Annand (Hemiptera: Adelgidae), has had well-documented negative impacts on eastern hemlock, *Tsuga canadensis* (L.) Carrière (Pinaceae), throughout much of the tree's native range in the eastern U.S. (Ellison et al. 2005; Eschtruth et al. 2006; Ford et al. 2012). Biological control has been a major component in management programs implemented against *A. tsugae* in forest systems (Reardon & Onken 2011; Havill et al. 2014). One of the primary species used in biological control programs directed against *A. tsugae* is *Laricobius nigrinus* Fender (Coleoptera: Derodontidae) (McClure 2000; Reardon & Onken 2011). *Laricobius nigrinus* is native to western North America, and since 2003 more than 200,000 adults have been released in the eastern U.S. (Havill et al. 2014).

Documentation of establishment and biology of *L. nigrinus* in areas of release has been crucial to biological control efforts; however, the use of traps to evaluate adult emergence and seasonal biology in the field is uncommon. Much of the research on seasonality and biology of *L. nigrinus* has involved field collections in areas of release using beat-sheets or branch clippings (Mausel et al. 2008, 2010; Hakeem et al. 2011), sleeve-cage studies in the field (Lamb et al. 2005, 2006; Mausel et al. 2008), or studies in the laboratory (Zilahi-Balogh et al. 2002, 2003a,b; Lamb et al. 2007). The use of traps may be an effective method to monitor *L. nigrinus* adults emerging from the soil following aestivation over the summer months, and enable land managers to assess population densities of this important predator. This method would provide more accurate population estimates in areas of release than many of the current means of sampling. Therefore, in 2010, a multi-year study was initiated to estimate densities of emerging adults of *L. nigrinus* based on emergence trap collections.

The study site was near Elkmont Campground, Great Smoky Mountains National Park (35.66417°N, 83.59028°W; about 640 m elevation; spanning the border between North Carolina and Tennessee), where 866 *L. nigrinus* adults were released on hemlocks at the edge of an open field on 14 Mar 2006. Establishment of *L. nigrinus* at this site was determined in 2009 (Grant et al. 2010).

To assess emergence of *L. nigrinus* in an area of release and establishment, emergence traps were placed beneath hemlock canopies at Elkmont. Traps consisted of a wood frame (4-sided pyramid shape,

77.5 × 77.5 × 50 cm, approximately 0.60 m² area per trap) covered with anti-virus screen (266 × 818 microns mesh size) (Green-Tek, Edgerton, Wisconsin), which guided beetles to a collecting head (120 mL unvented plastic specimen cup) (Lermer Plastics, Inc., Garwood, New Jersey) mounted to a wooden plate at the top of the trap (Wiggins et al. 2016). On 3 Oct 2010, 6 traps were placed under each of 4 canopies (tree heights ranged from about 7 m to about 14 m) of trees growing at the edge of an open field and monitored through 3 Mar 2011. Under each canopy, 3 traps were placed with the outer-most edge of the trap 1 m from the trunk (inner traps), and 3 traps were placed with the outer-most edge of the trap 2 m from the trunk (outer traps). The following season (on 6 Oct 2011), traps were moved to trees in the adjacent forest (within 50 m of the forest edge) to allow populations of *L. nigrinus* associated with previous study trees to grow, and have open ground on which to drop and develop in Spring 2012. Unlike Fall 2010, 3 traps were placed under each of 8 tree canopies, and traps were monitored until 1 Mar 2012. In Fall 2012, emergence traps were moved back to hemlock on the border of the field, and traps were placed in the same configuration under each of 4 trees as in Fall 2010. Traps were monitored from 26 Sep 2012 to 27 Mar 2013. During each sampling period in each sampling year, traps were monitored every 5 to 14 d.

Based on the total number of *L. nigrinus* adults that emerged under each tree each season, beetle density was estimated for each tree canopy by averaging the number of beetles per m² collected in traps under each canopy. Additionally, the area underneath each tree canopy was approximated by measuring the distance from the trunk to the dripline and calculating the area. The total number of beetles per tree canopy was estimated by multiplying the beetle density by the total area under each canopy.

Density estimates of beetles per m² from emergence trap collections at Elkmont ranged from 3.37 to 7.03 in 2010, 0.28 to 1.97 in 2011, and 1.41 to 14.90 in 2012 (Table 1). The extrapolated estimated total number of beetles per study tree ranged from 8 in 2011 to 658 in 2012. The average number of beetles estimated to occur beneath each study tree varied from 284 during 2010, to 35 during 2011, to 348 during 2012.

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Table 1. Total numbers of *Laricobius nigrinus* adults observed in emergence traps for each tree, and estimated numbers of adults per tree, Elkmont, Great Smoky Mountains National Park, 2010 to 2012.

| Year | Tree | Adults collected per tree (n) | Area under canopy (m ²) | Adults per m ² (n) | Estimated adults per tree (n) ^a |
|-------------------|------|-------------------------------|-------------------------------------|-------------------------------|--|
| 2010 ^b | 1 | 12 | 44.13 | 3.37 | 149 |
| | 2 | 18 | 73.48 | 5.06 | 372 |
| | 3 | 25 | 68.39 | 7.03 | 481 |
| | 4 | 14 | 33.94 | 3.94 | 134 |
| 2011 ^c | 1 | 2 | 102.98 | 0.56 | 58 |
| | 2 | 5 | 30.34 | 1.41 | 43 |
| | 3 | 7 | 29.17 | 1.97 | 57 |
| | 4 | 1 | 29.17 | 0.28 | 8 |
| | 5 | 7 | 23.63 | 1.05 | 25 |
| | 6 | 4 | 14.29 | 1.18 | 17 |
| | 7 | 2 | 14.29 | 1.12 | 16 |
| | 8 | 3 | 63.59 | 0.91 | 58 |
| 2012 ^b | 1 | 53 | 44.14 | 14.90 | 658 |
| | 2 | 18 | 73.48 | 5.06 | 372 |
| | 3 | 16 | 70.25 | 4.50 | 316 |
| | 4 | 5 | 33.95 | 1.41 | 48 |

^aCalculated estimates rounded and presented as whole numbers.

^bSix traps per tree.

^cThree traps per tree.

The numbers of *L. nigrinus* adults collected under canopies varied across years and may have been more similar in the 2010 and 2012 collections compared with the 2011 collections due to the location of trees sampled. Trees used during 2010 and 2012 were partially open grown and had fuller canopies, whereas trees during 2011 were in the forest or close to forest margins with less dense canopies. Partially open grown trees with fuller canopies provide more potential host material for *A. tsugae*, which, in turn, would facilitate greater numbers of *L. nigrinus* per tree. In some areas of release, greater numbers of *L. nigrinus* have been reported on release trees that are more exposed to direct sunlight (McDonald 2010). Despite differences in densities, emergence traps during all 3 yr were effective at collecting *L. nigrinus*. When compared with beat-sheet sampling conducted concurrently on the same study trees, traps collected similar numbers of *L. nigrinus* adults (Wiggins et al. 2016), with the added advantage of enabling population estimates. These estimates provide insight into the establishment and growth of *L. nigrinus* populations over time. The initial (and only) release of *L. nigrinus* at this site in 2006 was 866 individuals, and an estimated 1,394 adults were collected from 4 trees in 2012. Considering the numerous hemlock trees infested with *A. tsugae* in the area immediately adjacent to the 4 study trees, *L. nigrinus* populations have increased greatly since the initial release. Future use of emergence traps could enable the long-term evaluation of these populations and assist resource managers with collections of *L. nigrinus* for redistribution.

These results demonstrate that in addition to detecting *L. nigrinus* in areas of release and estimating its emergence period, emergence traps are useful for quantifying *L. nigrinus* populations in an area. Although beat-sheet sampling or branch clipping is effective at detecting *L. nigrinus* and estimating the seasonality of different life stages, the volume of a tree canopy is difficult to calculate, and therefore, the insect numbers based on that volume are difficult to extrapolate. The area underneath hemlock canopies is less difficult to estimate, and collections in a uniform unit area underneath the canopy allow for easier extrapolation of populations of *L. nigrinus*. This method can be used to estimate the number of insects per unit area for *L. nigrinus* and could be adapted for use with other biological control agents that have a life stage that emerges from the soil.

Summary

This study investigated the use of emergence traps to assess densities of *Laricobius nigrinus* Fender (Coleoptera: Derodontidae), a predator of hemlock woolly adelgid (Hemiptera: Adelgidae), in an area of release and establishment. Results indicate that traps effectively collect emerging adults and allow systematic population estimates of *L. nigrinus*. The incorporation of this method in future monitoring efforts could enable population estimates across the release range of *L. nigrinus*, thereby enhancing the evaluation of establishment and long-term persistence of this predator.

Key Words: *Adelges tsugae*; biological control; sampling; population estimation

Sumario

Este estudio investigó el uso de trampas de emergencia para evaluar la densidad de *Laricobius nigrinus* Fender (Coleoptera: Derodontidae), un depredador del pulgón de la tsuga (Hemiptera: Adelgidae), en un área de liberación y establecimiento. Los resultados indican que las trampas pueden capturar de manera efectiva a los adultos emergentes y permiten la estimación sistemática de la población de *L. nigrinus*. La incorporación de este método en los esfuerzos de vigilancia futuras podrían permitir a la estimación de la población en toda el área de liberación de *L. nigrinus*, mejorando de este modo la evaluación de establecimiento y persistencia a largo plazo de este depredador.

Palabras Clave: *Adelges tsugae*; control biológico; muestreo; estimación de la población

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