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Frontline

Forestry Research Applications

Canadian Forest Service – Great Lakes Forestry Centre

Technical No. 117

Use of ultraviolet light to assess hemlock woolly adelgid populations

The hemlock woolly adelgid (*Adelges tsugae* Annand, HWA) is a non-native invasive insect pest of eastern hemlock (*Tsuga canadensis* (L.) Carrière) in eastern North America. The HWA can often be found at the base of a needle on the underside of 1-yr-old twigs. This insect uses its mouthparts to pierce plant tissue and feeds on the tree's nutrient storage cells, reducing the tree's food reserves. Researchers at the Great Lakes Forestry Centre (GLFC) have developed a new method to use the by-product of this feeding as a way to obtain relative estimates of insect abundance.

INTRODUCTION

Estimates of insect abundance are one of the many factors used by managers when deciding if control treatments are needed or when evaluating the success of treatments. For HWA, estimates are obtained by collecting branches and counting the number of whitish-coloured ovisacs (a wool that covers nymphs, adults and eggs of HWA). However, not all ovisacs contain living HWAs, so the wool must be pulled apart to discern live from dead HWA to produce an accurate estimate of the population. Moreover, the density of ovisacs on twigs in spring can be very high and they often overlap, forming a loose mat of wool along the twig (Figure 1, left) that can make assessments time consuming and tedious.

When feeding, the HWA produces a waste product called honeydew, which appears as yellow drops of fluid on the exterior of ovisacs (Figure 1, left). Research in the United States[1] has shown that both ovisacs and honeydew glow when exposed to ultraviolet light (UV) (Figure 1, right). This work prompted researchers at GLFC to study the relationship between the number of honeydew droplets and counts of live HWA. If these two factors are related, estimates of HWA population levels could be obtained simply by counting honeydew droplets under UV instead of dissecting ovisacs. This technique is expected to save considerable time estimating HWA populations.

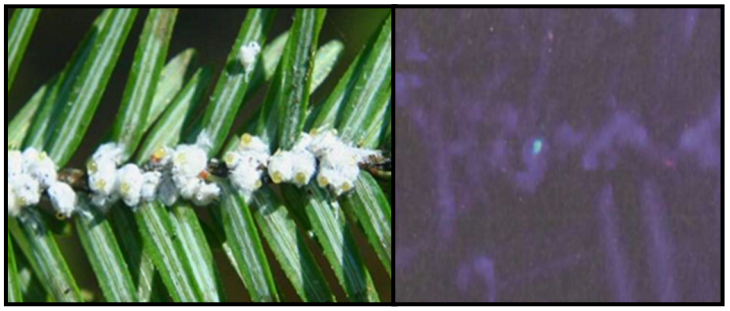

Figure 1. Left: A hemlock twig with densely packed ovisacs of maturing HWA (Tom Coleman, US Forest Service, Bugwood.org). Honeydew can be seen as tiny yellowish droplets on the surface of the ovisacs; Right: Bluish-white colour of a honeydew droplet on an undisturbed HWA ovisac[1] under UV.
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Methods

Branch samples were collected in early April, 2014 in upper New York State and in late April-early May, 2018 in southwestern Nova Scotia (where HWA was discovered in 2017). Ovisacs were small (1-2 mm diameter) and appeared to be at low density on twigs in 2014 and larger (2-4 mm diameter) and of higher density on twigs in 2018. In fact, ovisacs had grown and coalesced with neighbouring ovisacs on many twigs in 2018. The steps for assessing these branch tips are provided below (see "**Procedure**"). The strength of relationship between UV and dissection counts was examined using regression analysis. Once each model was developed and assessed, it was also validated with data that was excluded from the regression analysis.

Findings

Honeydew droplets were easily seen and counted under UV and were related to counts of live HWA (Figure 2). In the 2014 collection, branch tips contained 0-22 live ovisacs and the relationship was very strong. In the 2018 collection, tips contained many live ovisacs (0-255) and the relationship was weaker than 2014. In 2018, to speed assessments, the larger

[1] McDonald, R.C.; Kok, L.T. 2014. A simple method of detecting hemlock woolly adelgid (Hemiptera: Adelgidae) predator activity using ultraviolet-A light. Journal of Entomological Science, 49(2): 200-205.

ovisacs were not dissected but instead were assumed to contain living HWA so they were counted 'as is'. Assessment of branch tips in 2014 with UV took ~1 minute per 30 cm tip whereas dissection of the same tips took ~10 minutes.

There are, however, some limitations with this technique. First, not all live nymphs produced honeydew during the exposure time at room temperature. A simple solution to this would be to hold branch tips longer at room temperature to give HWA more time to produce honeydew and for it to reach the ovisac's surface. Second, the UV technique might underestimate the number of live HWA if ovisacs have formed a loose mat. This is because the honeydew appears to diffuse through the wool rather than emerging on the exposed surface of the ovisac. If assessment of twigs with high density of ovisacs is necessary then either a different technique, such as ovisac dissection, should be used or, when reporting the data, acknowledge that the accuracy of the estimates made with UV might be degraded.

We suspect that the relationship found in 2018 could be improved. For example, not all ovisacs were dissected from this collection and, as described above, a difference in ovisac size is no guarantee that the HWA within it is alive or dead. Based on our experience we advise that before implementing a sampling scheme based on this method users should apply the procedures listed below to develop their own models. This technique should not be used after ca. half of the overwintering generation (sistens) has begun laying eggs (typically February to May). We suspect that HWA produces very little to no honeydew during this period but more work is needed to confirm these observations. This method should not be used to estimate the density of the summer generation of HWA (progrediens) because these individuals grow among the sistens ovisacs and quickly become coalesced with neighbours.

Our results show that when HWA is at low density, counts of honeydew droplets made under UV can save significant time compared to dissections. Adopting this method will therefore shorten the time needed to process samples. On the other hand, this technique makes possible the assessment of additional branches in the same amount of time needed for dissections, which could increase the accuracy of population assessments.

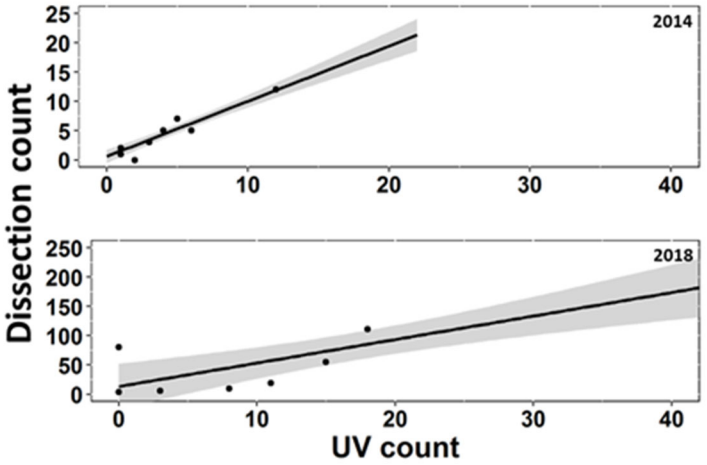

Figure 2. Relationship between counts of honeydew droplets reflecting UV light (x) and of live HWA during dissections (y) of branch tips from New York in 2014 and Nova Scotia in 2018. Shaded area is the standard error of the models. Round symbols show how well additional samples collected from each site corresponded to the fitted models (see text for details). Equation for all 2014 data: y = 0.47 + 0.96x; for all 2018 data: y = 12.76 + 3.96x.

PROCEDURES

The following steps should be followed to replicate our results.

1. Collect branch samples with ovisacs any time from fall through spring (September to February-May depending on region).
2. Keep branches at 4°C for a few days before assessments. This will slow HWA feeding.
3. When ready to process samples, transfer each branch from storage to room temperature and place the cut end in a bucket of warm water (10-15 cm deep) for 1-2 hours before beginning assessments.
4. Remove a branch from the water and trim off a 30-cm branch tip.
5. Place each branch tip upside down on a bench in a dark room.
6. Hold a UV light within 15 cm and directly overhead of the branch and also at a 45° angle from the tip, the base and the two sides.
7. Count the number of honeydew droplets reflecting the UV (Figure 1; right).

To build a new model or to add to those in Figure 2, ovisacs must be dissected and the number of live HWA counted. To do this:

1. Place the branch tip under a microscope with 64× magnification.
2. Pull apart the wool of each ovisac or group of ovisacs to reveal each HWA.
   a. Nymphs and adults that that are alive will appear reddish-brown and plump; the rest are dead. If unsure, gently touching the HWA will cause live individuals to emit honeydew or move their stubby legs. HWA that appear dead but have eggs present should be counted as alive as the eggs are viable.

3. Record the number of live nymphs, adults and egg masses.
4. Plot the relationship between the UV count of honeydew and the dissection counts as in Figure 2 and compute the regression relationship.

HAND-HELD *UV* LIGHTS (315-400 NANOMETRES WAVELENGTH) WERE USED TO COUNT HONEYDEW DROPLETS. THESE UNITS ARE READILY AVAILABLE FROM FORESTRY OR ENTOMOLOGY SUPPLY STORES (~$50).

CONCLUSION

While local correlations need to be determined by each user, this method is relatively inexpensive and has potential for reducing the time required to obtain population estimates, as well as improving overall accuracy. It is another useful tool in managing HWA infestations. For further information on HWA, refer to Frontline Technical Notes 114 and 116.

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COLLABORATORS

Cornell University (Mark Whitmore)

Canadian Food Inspection Agency (Ron Neville)

Invasive Species Centre

Nova Scotia Department of Lands and Forestry (Jeffrey Ogden, Celia Boone)

Ontario Ministry of Natural Resources and Forestry

USDA Forest Service

New York State Department of Environmental Conservation

CONTACT INFORMATION

Jeffrey G. Fidgen, Chris J. K. MacQuarrie, Jean J. Turgeon
Great Lakes Forestry Centre
1219 Queen St. East
Sault Ste. Marie, Ontario, Canada
P6A 2E5
Phone: 705-949-9461
E-mail: nrcan.glfcwebglfc.rncan@canada.ca
Web site : nrcan.gc.ca/forests/research-centres/glfc/13459

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