Western Hemlock Looper

Lambdina fiscellaria lugubrosa (Hulst)
Lepidoptera: Geometridae

Shepherd, R. F.; Gray, T. G. 1972. Solution separation and maximum likelihood density estimates of hemlock looper (Lepidoptera: Geometridae) eggs in moss. Canadian Entomologist 104: 751-754.

Objective: To describe an efficient method of processing *L. fiscellaria lugubrosa* eggs in moss samples and determining egg density.

Abstract: Western hemlock looper, *Lambdina fiscellaria lugubrosa* (Hulst), is an important defoliator of western hemlock, *Tsuga heterophylla* (Raf.) Sarg., and other conifers in the USA and Canada. Periodical damage generally occurs in mature or senescing stands, where defoliation results in growth reduction, top kill, and tree mortality. Eggs are laid on moss, bark, and organic debris on the trunks and branches of trees and on the ground. Visual surveys of eggs in moss samples are tedious and often inaccurate as not all eggs are easily seen and empty chorions can be mistaken for viable eggs. A two-step separation technique using solutions of NaOH and NaCl was developed to process *L. fiscellaria lugubrosa* more efficiently. Eggs obtained in this manner can be used to determine egg density per unit of sampled moss or duff. Collected eggs can be reared if the NaOH soak is skipped. The entire process of soaking, rinsing, and separating a sample took an average of 15–20 minutes, not including time spent counting eggs on the filter paper.

The maximum likelihood method of sampling (MLM; Southwood 1966) can be used to determine the density of eggs in each moss sample. The MLM assumes that a constant proportion of eggs will be removed from the sample with each wash, but removing all eggs is not necessary. This method requires that each wash in the separation technique take exactly 2 minutes with the same amount of effort for each wash in each sample. This technique was evaluated with 35 464 cm^2 samples of moss and was considered to be efficient in estimating egg densities. Assuming a population of 200 eggs in each sample, 75% of the eggs must be collected by the three washes to obtain a coefficient of variation of ≤10%. This was exceeded in that the technique removed an average of >98% of the eggs in each sample.

Sampling Procedure: Collect 464.5 cm^2 samples of moss from the bark surface of hemlock trees. Soak each sample for one minute in separate containers filled with 0.5% NaOH solution. For each container, pour the contents onto a set of circular mesh screens, 20 cm wide, with the top and bottom screens having 1.00- and 0.354-mm mesh openings, respectively. Rinse the container with water and pour onto the mesh as well. Mix the moss on the upper screen continuously for exactly two minutes while irrigating the material with water from a hose at full tap pressure. Separate the two screens and wash the eggs and other fine debris off the bottom screen into a collection jar. Join the two screens again and wash the moss sample on the upper screen for exactly two minutes twice more, washing the eggs and other debris into

the collection jar each time. Pour the contents of the collection jar onto a 12.4-cm disk of filter paper set in a Buchner vacuum funnel. Vacuum filtrate the contents, then count the number of eggs on the filter paper using a stereomicroscope.

Samples of moss or duff under trees require differential floatation to separate *L. fiscellaria lugubrosa* eggs from the very fine organic debris. After vacuum filtration, use a 15% NaCl solution to wash the eggs and fine material off the filter paper and into a 500-ml separatory funnel. Add enough 15% NaCl solution that the contents can mix easily and shake the funnel well. Intact eggs should float to the surface while empty chorions and other extraneous material sink to the bottom. Loosen the stopcock to remove this material from the funnel. The upper solution with intact eggs can be poured onto a 12.4-cm disk of filter paper set in a Buchner vacuum funnel. Rinse with additional NaCl solution and pour the rinsate onto the filter paper. Vacuum filtrate the contents, then count the number of eggs on the filter paper using a stereomicroscope. The NaCl solution can be reused for subsequent separations.

Note: Exposures of longer than one minute to the NaOH soak will dissolve the chorions and degrade the eggs, leading to inaccurate counts of egg densities.

Reference:

Southwood, T. R. E. 1966. Ecological methods with particular reference to the study of insect populations. London: Methuen; 391 p.