A PROCEDURE FOR SAMPLING NYMPHS OF SARATOGA SPITILEBUG, *APHROPHORA SARATOGENSIS* (HOMOPTERA: CERCOPIDAE), USING PERCENTAGE OF SAMPLE-UNITS INFESTED

Louis F. Wilson and Sharon L. Hobrla

**ABSTRACT**

A method is proposed for rapidly estimating the nymphal population of the Saratoga spittlebug on alternate host plants in young red pine plantations. The method is based on an assumption that the number of nymphs per sample unit is distributed within an infested plantation according to the negative binomial distribution. This method can be 1.1–11.0 times more efficient depending upon the density of plants and the nymphal population level.

The Saratoga spittlebug, *Aphrophora saratogensis* (Fitch), is one of the most destructive pests in young pine plantations in eastern North America. Adult spittlebugs feed on the branches of pine, preferring red pine, *Pinus resinosa* Aiton, planted in fields among abundant alternate hosts, especially sweet-fen, *Comptonia peregrina* (L.) Coult. Heavy feeding drastically reduces shoot growth and can badly deform or kill trees if repeated for several years (Ewan 1958, Wilson 1987).

The traditional method of surveying for spittlebug nymphs devised by Ewan (1958, 1961) is still being used today, with minor variations. Conducted in the spring, it requires using a square 0.1-milacre sampling frame (4,225 cm²) placed on the ground. All nymphs are counted on the alternate hosts in 50 or more sample frames taken throughout one or more specified units (usually 0.1-acre plots) of the pine plantation. Although such sampling provides a reliable estimate of the nymphal population in high- or moderate-risk areas of a stand, this method requires much time to locate and count nymphs in their spittlemasses. Occasionally 50 or more nymphs may be present in a single sample, and the sampling rules require the surveyor to continue counting until all nymphs are tallied. Counting small nymphs while on one's hands and knees is tedious, and searching for them may take more than half the time allotted for the survey.

To alleviate the drudgery of counting nymphs, we propose a faster method for estimating the nymphal population. The new procedure is based on the assumption that the number of nymphs per sample unit is distributed within a red pine plantation according to the negative binomial, so that an estimate of the mean number of nymphs per sample-unit may be derived by determining the proportion of the sample-units containing nymphs. In this paper, we develop this technique and discuss its time-saving feature.

**MATERIALS AND METHODS**

This study was made in three sapling-size red pine plantations designated A, B, and C that were located in Traverse, Lake, and Alcona counties in Michigan. Thirteen 1/5-acre

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Table 1. Statistics computed from individual Saratoga spittlebug nympha counts from each red pine plantation (PL) each year.

<table>
<thead>
<tr>
<th>Pl.</th>
<th>Date</th>
<th>No. of plots</th>
<th>No. of samples</th>
<th>Proportion infested</th>
<th>Mean count</th>
<th>Variance</th>
<th>( \bar{X} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>01/6/72</td>
<td>5</td>
<td>250</td>
<td>0.068</td>
<td>0.092</td>
<td>0.020</td>
<td>0.568</td>
</tr>
<tr>
<td>A</td>
<td>31/5/73</td>
<td>4</td>
<td>200</td>
<td>0.090</td>
<td>0.105</td>
<td>0.027</td>
<td>2.431</td>
</tr>
<tr>
<td>C</td>
<td>30/5/73</td>
<td>4</td>
<td>200</td>
<td>0.145</td>
<td>0.195</td>
<td>0.066</td>
<td>0.773</td>
</tr>
<tr>
<td>B</td>
<td>28/6/71</td>
<td>5</td>
<td>175</td>
<td>0.172</td>
<td>0.424</td>
<td>0.222</td>
<td>0.323</td>
</tr>
<tr>
<td>B</td>
<td>31/5/72</td>
<td>5</td>
<td>250</td>
<td>0.200</td>
<td>0.364</td>
<td>0.125</td>
<td>0.626</td>
</tr>
<tr>
<td>A</td>
<td>29/6/71</td>
<td>5</td>
<td>175</td>
<td>0.211</td>
<td>0.450</td>
<td>0.268</td>
<td>0.364</td>
</tr>
<tr>
<td>A</td>
<td>04/6/71</td>
<td>5</td>
<td>125</td>
<td>0.216</td>
<td>0.376</td>
<td>0.149</td>
<td>0.582</td>
</tr>
<tr>
<td>C</td>
<td>30/5/72</td>
<td>3</td>
<td>150</td>
<td>0.220</td>
<td>0.460</td>
<td>0.939</td>
<td>0.213</td>
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<tr>
<td>B</td>
<td>04/6/71</td>
<td>5</td>
<td>125</td>
<td>0.272</td>
<td>0.648</td>
<td>0.583</td>
<td>0.361</td>
</tr>
<tr>
<td>A</td>
<td>05/6/71</td>
<td>5</td>
<td>250</td>
<td>0.324</td>
<td>0.836</td>
<td>1.026</td>
<td>0.368</td>
</tr>
<tr>
<td>B</td>
<td>30/5/73</td>
<td>5</td>
<td>250</td>
<td>0.396</td>
<td>0.792</td>
<td>0.245</td>
<td>2.151</td>
</tr>
<tr>
<td>A</td>
<td>08/5/75</td>
<td>5</td>
<td>250</td>
<td>0.428</td>
<td>1.680</td>
<td>1.821</td>
<td>0.675</td>
</tr>
<tr>
<td>C</td>
<td>28/6/71</td>
<td>3</td>
<td>105</td>
<td>0.476</td>
<td>1.390</td>
<td>1.703</td>
<td>1.015</td>
</tr>
<tr>
<td>B</td>
<td>03/6/74</td>
<td>5</td>
<td>250</td>
<td>0.500</td>
<td>1.848</td>
<td>3.566</td>
<td>0.574</td>
</tr>
<tr>
<td>C</td>
<td>03/6/74</td>
<td>3</td>
<td>150</td>
<td>0.633</td>
<td>1.720</td>
<td>4.383</td>
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</tr>
<tr>
<td>C</td>
<td>02/6/71</td>
<td>3</td>
<td>75</td>
<td>0.707</td>
<td>2.080</td>
<td>4.847</td>
<td>1.428</td>
</tr>
<tr>
<td>C</td>
<td>29/5/75</td>
<td>3</td>
<td>150</td>
<td>0.713</td>
<td>4.780</td>
<td>49.807</td>
<td>0.708</td>
</tr>
<tr>
<td>B</td>
<td>29/5/75</td>
<td>4</td>
<td>200</td>
<td>0.745</td>
<td>6.505</td>
<td>68.975</td>
<td>0.746</td>
</tr>
</tbody>
</table>

(0.08 ha) circular plots were set up among the three study areas, and the nymphs were counted yearly in each plot in the spring from 1971 to 1975. Nymphal counts were taken twice (one month apart) in a few plots to check on nymph feeding locations and mortality. Due to the rapid population fluctuations of the spittlebugs, repeated measurements in the same plantations showed no significant correlations. In all, 77 plot data sets were acquired. Nymphal samples were taken randomly using a standard square frame (65 cm x 65 cm) used for spittlebug surveys. Sample means were calculated from 35 to 50 frame counts. Ewan (1961) showed that an area of 4,225 cm$^2$ (the frame) was a highly efficient sample-unit size and as reliable as comparable units 10 times as large. The plots encompassed a wide range of infestation levels so that means for the individual 77 plot data sets varied from 0.02 to 12.32 nymphs per sample frame. The statistics computed from the data for each plantation by sample date are summarized in Table 1. The overall statistics in the table were calculated from the 3,330 individual sample-unit (frame) data.

To estimate the time saved by the new method, we compared it to the traditional one using the following assumptions:

1. The mean number of nymphs expected in a plot of 50 samples ranges from 1 to 13 nymphs per frame. We set the upper limit at 13 because this just exceeded the mean number found in one heavily infested plot in this study. One nymph was set as the lower limit because a few test calculations suggested that at numbers lower than one the nymphs would be so scarce that there would be little difference in counting time between methods.

2. The mean number of alternate hosts per sampling frame is 42 plants with a range of 29–74. All suitable alternate hosts were counted in 650 sample-units from the 13 sample plots in this study, but only the 10 plots that we ranked moderate or high risk for spittlebug were used. In normal surveys, low-risk areas of a stand would not be sampled for nymphs. The 500 sample-frames averaged 38 (range 19–66) forbs and four (range 1–10) sweet-fern plants. Sweet-fern was separated from the forbs in the analyses because the older nymphs congregate on sweet-fern, so that after mid-June, 60–80% of the insects are on this host
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![Graph showing negative binomial distribution fitted to mean number of Saratoga spittlebug nymphs per sample-unit on the proportion of sample-units infested. Confidence intervals for sample sizes 50, 100, and 1000 are at the 95% level.

Fig. 1 Negative binomial distribution fitted to mean number of Saratoga spittlebug nymphs per sample-unit on the proportion of sample-units infested. Confidence intervals for sample sizes 50, 100, and 1000 are at the 95% level.

(Ewan 1961). In the new method the surveyor examines sweet-fem first because of the better chance of finding nymphs there.

3. The time to search each plant averages 3 sec. and to count each nymph 5 sec. This means that in the traditional method surveyors might occasionally count 50 nymphs on some plants (250 sec.), but in the new method they would never count more than one nymph (for a maximum of 5 sec.). These times were estimated but considered reasonable based on several years of field experience with spittlebug sampling by one researcher and one technician.

The time calculations were made in terms of 50 sample-units, the number taken in a sample plot, by considering combinations of high and low insect populations and high and low plant densities using the assumptions given. The calculations considered the proportion of the frames infested and those not infested, data obtained from Figure 1. In a laboratory simulation of the traditional method, we examined all plants and counted all insects in the infested frames. Similarly, in a simulation of the new method, we first examined sweet-fem with a 70% probability of finding a nymph on one out of four plants. In the instances that nymphs were only on forbs and not on sweet-fem, we assumed the surveyor would have to examine one third of the forbs to locate a nymph. For the frames that were not infested, we examined all plants with both methods. The calculations for each method, expressed in minutes per plot, are compared in Table 2.
Table 2. Times calculated to sample 50 frames (one plot) for Saratoga spittlebug nymphs and time saved at different nymphal population levels and plant densities.

<table>
<thead>
<tr>
<th>Nymphal population</th>
<th>Plant density</th>
<th>Minutes per plot</th>
<th>Time saved</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Traditional</td>
<td>New</td>
</tr>
<tr>
<td>Low</td>
<td>High</td>
<td>127</td>
<td>116</td>
</tr>
<tr>
<td>Low</td>
<td>Low</td>
<td>77</td>
<td>47</td>
</tr>
<tr>
<td>High</td>
<td>High</td>
<td>239</td>
<td>41</td>
</tr>
<tr>
<td>High</td>
<td>Low</td>
<td>189</td>
<td>17</td>
</tr>
</tbody>
</table>

THE ESTIMATION PROCEDURE

The negative binomial distribution, a more general form of the Poisson, includes a parameter $\kappa$, which is an index of dispersion or aggregation (Anscombe 1949). It is represented by the function

$$\text{Prob}(Y = y) = \left(\frac{\kappa + y - 1}{y}\right) \left(\frac{\kappa}{\lambda + \kappa}\right)^\frac{\kappa}{\lambda + \kappa} y$$

Wilson and Gerrard (1971) showed that the negative binomial distribution is well suited for estimating population levels of insects such as the European pine sawfly, *Neodiprion sertifer* (Geoffroy), in young pine plantations. An important feature of the negative binomial model is its ability to account for a contagious or clumped distribution, a characteristic of insect and most other biological populations. Mate seeking, host selection, feeding preference, oviposition, and other congregating habits tend to result in more insects at some locations and less at others. Under the assumption that spittlebug nymphs are more likely to occur in this clustered pattern, the negative binomial is an appropriate model of their population distribution.

The estimation model for the negative binomial distribution can be written in the form

$$\hat{\kappa} = \kappa \left[\left(\frac{1}{1 - \pi}\right)^{1/\kappa} - 1\right]$$

where $\lambda$ is the estimated number of spittlebug nymphs per 0.1-milacre given $\pi$, $\pi$ represents the proportion of sample frames expected to contain at least one nymph, and $\kappa$ is an index inversely proportional to the propensity of the nymphs to aggregate.

The first step in the analysis required that we find an appropriate estimate of $\kappa$. Using half-interval functional minimization (Carnahan and Wilkes 1973), the estimate was 0.750; and when applied to the spittlebug data, it translated into the curve shown in Figure 1. The curve fits the data with an $R^2 = 0.925$ and a standard error of the residuals about the regression of 0.538.

The variance was calculated as recommended by Wilson and Gerrard (1971) using

$$\hat{V}(\hat{\kappa}) = \left[\frac{1 + \ln(1 - \pi)}{\hat{\kappa}^2}\right] \left[\kappa + \ln(1 - \pi)\right] - 1$$

Approximate 95% confidence intervals for samples of 50, 100, and 1000 are shown on the graph in Figure 1.
A test of significance between $k$ and density produced an F-statistic of 0.28, supporting
the assumption of a $k$-estimate that was independent of population density.

SURVEY TIMES COMPARED

The time-saving calculations indicated that the new method is faster than the traditional
method. However, it is only 1.1 times faster when the insect population is low and plant
density is high (Table 2). This is the worst-case situation, one which might be expected
because in both methods, numerous plants must be examined before an insect is located,
and many frames would have no insects requiring the checking of all plants. With the
assumptions given for this situation, we calculated 127 min. for sampling by the
traditional method versus 116 min. by the new method (Table 2).

In the contrasting situation, where the insect population is high and plant density is low,
the time saved is more than 11 times. In this instance a plot of 50 frames can be sampled
in 17 min. or less, whereas the traditional method required more than 3 h. (i.e., 189 min.)
(Table 2). Actually in this case, even more time might be saved by the new method. The
reason for this is that where there is a high insect population, the surveyors can readily
spot a large spittlemass on a plant even at some distance. For all practical purposes, they
can tally that frame as infested without directly examining the contents of the spittlemass.

The shortest sampling time for the traditional method occurs when both the nymphal
population and the plant density are low. We calculated 77 min. for sampling, but with
the new method the time is still shorter by 30 min. for a 1.6 times more rapid sample
(Table 2).

Where both the nymphs and plants are numerous, sampling takes the longest time when
using the traditional method. Admittedly this is an extreme case, so that counting nymphs
and searching plants by the traditional method took nearly 4 h. (239 min.). The new
method reduced sampling to 41 min. for a survey that was nearly 6 times faster (Table 2).

DISCUSSION

The negative binomial predicted the nymphal population within limits suitable for
developing a new and more efficient spittlebug sampling procedure. Sampling the
spittlebug by tallying the presence or absence of nymphs, instead of counting all nymphs
in each frame as in the traditional method, saves time with little loss in the accuracy of
estimation. The time saved, however, varies considerably depending on the size of the
nymphal population and density of the alternate-host plants, the two primary variables
concerned with nymphal sampling. For example, when there are many nymphs, sampling
is rapid by the new method and particularly slow by the traditional method. But, the speed
is further mitigated by the number of plants the observer has to check. If there are
numerous plants, sampling generally takes longer in both methods, but in the new method
the surveyor can quickly overview the plants to locate a spittlemass instead of
systematically starting from the corner and checking all plants. Often, spittlemasses can
be seen at some distance, especially when the nymphs congregate on sweet-fern after
mid-June.

Recent research indicates that all nymphal sampling should be done after mid-June for
several reasons (Wilson 1987). The young nymphs (1st-2nd instars) are difficult to locate
because they are small, and about 80% are widely dispersed on the forbs (Ewan 1958).
After mid-June, the nymphs are larger and there is a high probability that the nymphs will
be on sweet-fern where their spittlemasses are easier to locate. The primary reason for late
sampling, however, is that the population of large nymphs correlates well with the adult
population and thus predicts adult spittlebug damage with greater accuracy.

The new survey procedure changes only the way of sampling within the frames and not
the other aspects of the sampling technique. However, by the traditional method nearly
two thirds of the time of sampling involves searching plants and counting nymphs. So with the new method, the entire sampling technique can be reduced by one third or by one half when the nymphs are abundant.

LITERATURE CITED


