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Gypsy Moth Egg Mass Sampling for Decision-Making: *A Users' Guide*

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Since the time of its accidental introduction to North America in 1868 or 1869, the gypsy moth, *Lymantria dispar* (L.), has become one of the most destructive forest insects in the northeastern U.S. In the wake of the gypsy moth's continuously expanding infested front, over 40 million acres were defoliated over the last 10 years. Though little can be done to stop the spread of this insect, considerable resources are expended to suppress outbreak populations and reduce the rate of spread. Under the U.S. Cooperative Forestry Assistance Act of 1978, over 7 million acres of forest land in the United States have been sprayed with insecticides to suppress this insect during the past decade.

A key element of any forest pest management program is the careful evaluation of a forest stand to determine whether population levels indicate that intervention is appropriate. The two most commonly used measures of gypsy moth density are counts of males in pheromone traps and counts of over-wintering egg masses. Pheromone traps are mostly used in isolated populations outside of the generally infested area and in areas along the expanding front of the gypsy moth infestation. In these areas, pheromone traps are useful for detecting and delineating new infestations and for identifying rising populations that warrant more intensive surveys. Within the generally infested area, where management program goals are usually to suppress populations or prevent defoliation, counts of over-wintering egg mass populations are relied upon to make decisions concerning control. The availability of egg masses from late summer through spring provides ample time for censusing population levels while still leaving adequate opportunity for treatment planning. Furthermore, gypsy moth egg masses are probably the most convenient stage to sample because most are visible from the ground.

As the range of the gypsy moth expands, there is a need to educate forest pest management personnel and others on methods available for censusing egg mass populations. Furthermore, there have been some important scientific contributions to the development of better approaches to egg mass census methodologies, and there is a need to communicate these findings to users. In this handbook we provide a "how-to" description of procedures to use for censusing egg mass populations, and also provide information on how these data should be used in decision-making.

The objective of most gypsy moth egg mass sampling efforts is to estimate egg mass density, expressed as number per unit ground area (usually number per acre). Egg mass densities are typically expressed as numbers per unit area because ground area is a stable unit of measure. Expression of densities as numbers per tree is less desirable because the number and size of trees per acre varies from stand to stand and egg masses are often not located on trees. Density estimates can be used to predict defoliation levels using one of several previously developed density-defoliation relationships (see page 15). Most management programs simplify matters further by establishing density thresholds for invoking intervention; these egg mass density thresholds correspond to impact thresholds, above which defoliation and other impacts are considered intolerable.

Probably the simplest method for censusing egg mass densities is with the use of "fixed-radius" plots. Under this method, all of the egg masses within a circle (usually covering 1/40th acre) are counted (see page 8 for a detailed description of this method). Several of these plots are replicated throughout the stand in order to estimate density. This technique has the advantage of simplicity; density is simply the average density from all plots.

Another commonly used approach is the "fixed- and variable-radius" plot technique. Under this procedure, all the egg masses are counted on trees selected from the plot center using a prism, a tool commonly used by foresters to estimate tree basal area. The prism is used to determine which trees are inside or outside of the plot. Egg masses occurring on objects other than trees (such as fallen trees and branches) are counted within a fixed-radius plot located around the same plot center. These plots are then replicated through out the stand to estimate density. While this procedure provides density estimates comparable with those obtained using the fixed-radius plot approach, it is substantially more complicated and consumes more time. We recommend the use of fixed-radius plots because they provide the same level of precision at less cost.

A third technique for estimating egg mass densities that has been widely used is the five-minute walk. Under this procedure, an individual walks randomly through the stand for five minutes, counting all egg masses that are readily observable. These walks are typically replicated by two or more individuals and the average counts are then converted to estimates of egg mass density (number per acre) using previously developed tables of equivalency. The statistical properties of this procedure have recently been examined in detail. These reports recommend against this procedure for two reasons: 1) density estimates vary considerably among counts taken by different observers and 2) even when just one person performs the counts, these counts cannot be converted to density estimates with adequate precision.

Fixed-Radius Plot Method - Procedures

Survey area The first step in conducting a gypsy moth egg mass survey is to identify the boundaries of the potential treatment block over which to estimate egg mass density. This may be simple as in the case of an isolated, uniform oak woodlot. It may be more complicated in areas that are not continuously wooded or in wooded areas in which the density of preferred tree species is not uniform. In the latter situation, it may be advisable to break the total area up into smaller areas for the purposes of estimating egg mass density. The treatment block boundaries should be located such that each unit is relatively homogenous both in terms of management objectives and in terms of forest composition. Examination of egg mass data may indicate that boundaries should be redrawn to exclude areas with low egg mass densities. Treatment blocks should range from 10 to 500 acres; any area larger than 500 acres may be so heterogeneous that it should be subdivided.

Number of Samples Gypsy moth egg masses are typically irregularly distributed within a stand. For example, large gnarly oak trees tend to accumulate large numbers of egg masses. Plots containing one of these so called 'wolf ' trees would provide a much higher density estimate than would an adjacent plot without such a tree. It is this type of situation that causes the variation between plots and creates the need to sample using several plots for each potential treatment block.

The number of sample plots that are necessary is determined by the expected variability among samples and by the maximum estimation error that is acceptable. This error is often expressed as a proportion of the estimated density. The maximum estimation error depends upon the constraints of the management program and the density one is estimating. For example let us consider an estimation error of 50%; if the estimated density is 4,000 egg masses per acre, the manager may not care whether the actual density is 2,000 or 6,000 egg masses per acre, since treatment would be required even at 2,000. In this case an error of 50% would be acceptable. But if the estimated density is 400 egg masses per acre and the treatment threshold is 500 egg masses per acre, the decision whether to treat may be highly dependent on whether the actual density is 200 or 600 egg masses per acre. In this case an estimation error of 50% would be unacceptable.

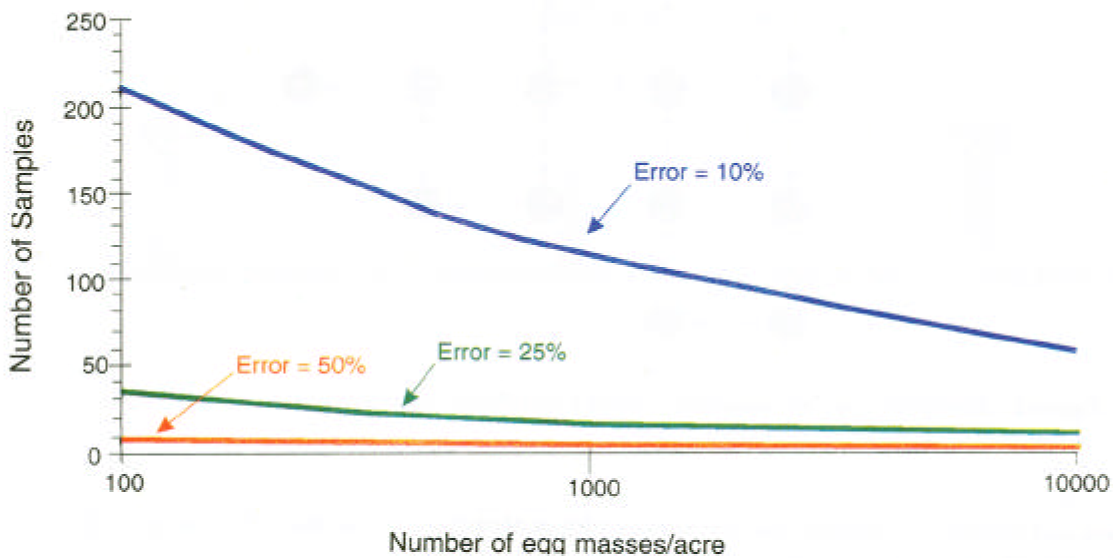

Fig. 1. Minimum number of fixed radius samples (plots) necessary to achieve various levels of precision at different densities. Error is expressed as a percentage of estimated density.

The required number of samples is also dependent upon the expected variability among samples. Typically, the variability among samples is strongly related to gypsy moth density: variation is greatest at high densities. Figure 1 depicts curves which give the minimum number of samples required at different population densities for a variety of maximum errors[5]. To use these curves requires two pieces of information: 1) the maximum allowable error and 2) an estimate of density. When using this graph the estimate of density (horizontal axis) can be a very approximate value obtained from a superficial examination of the stand.

If an error of 25% is considered acceptable, and the population density is expected to be greater that 2000 egg masses per acre, then 15 samples are probably adequate. However, nearly 100 samples are needed to achieve a 10% error rate at the same population density. Under low density conditions, (<500 egg masses per acre) up to 33 samples are needed to maintain a 25% error rate, and over 200 sample are required for a 10% error rate.

Location of survey points Sampling theory tells us that samples should be placed randomly throughout the management unit. However, it is usually simpler, and still acceptable, to space the survey plots evenly. An easy way to do this is to draw a grid of lines on a map of the management unit as a guide for drawing the locations of the desired number of survey points (Fig. 2). The grid points can be located by starting at some easily located landmark (e.g., a road) and pacing out distances between sample locations while following a fixed compass direction.

[5]These values are based on samples from 35 acre tracts in central Pennsylvania, and are calculated using a 75% confidence interval. In other words, at a given population density, one can be 75% certain of achieving the indicated error rate if the indicated number of samples are taken.

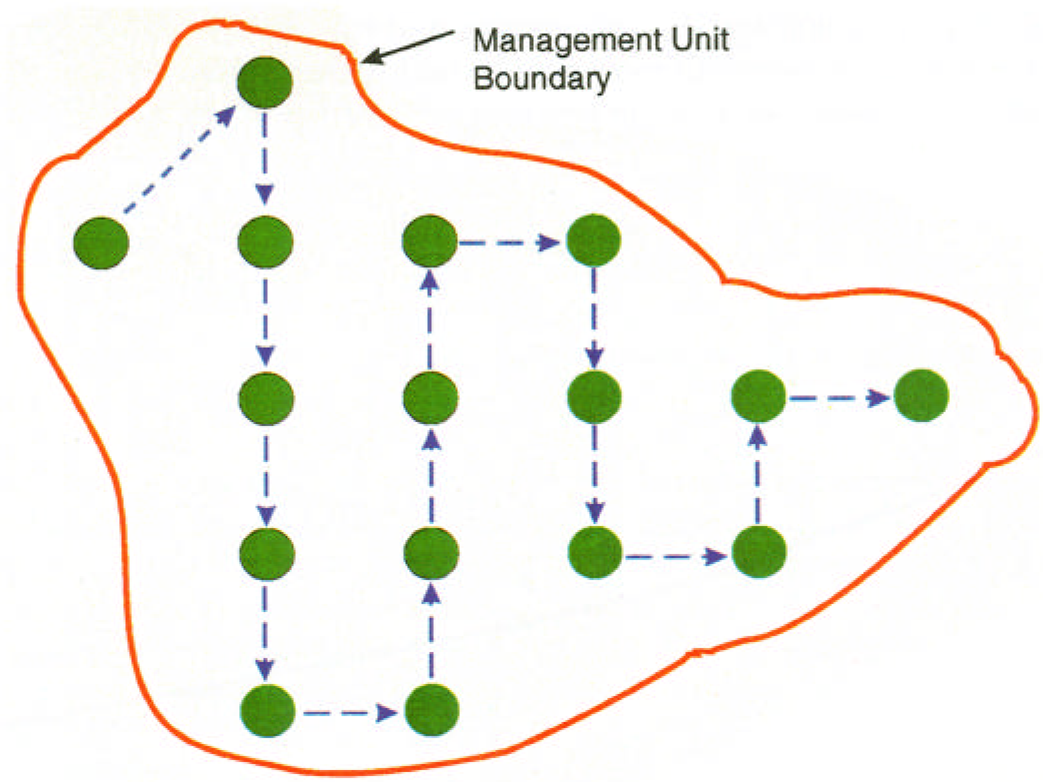

Fig. 2. Arrangement of fixed-radius plots within a hypothetical management unit (efficient routes for walking between plots are shown as dashed lines).

The exact location of the center of the survey plot is of critical importance, as it can greatly influence the number of egg masses that will be counted in that plot. Gypsy moth egg masses have a highly clumped distribution, which means that two side-by-side survey plots can contain vastly different numbers of egg masses. Since it is possible to learn to recognize those locations which will tend to contain the most egg masses, it is also possible to deliberately place survey points in these locations (or to deliberately avoid these locations). Either way, the estimated egg mass density for the management unit will not be accurate if such deliberate adjustments are made. In statistical terms, this is known as "bias". Deliberately locating survey plots in areas where egg masses are clumped would bias the estimates upward, resulting in an overestimate of the true egg mass density for the management unit. Deliberately locating plots away from clumps would bias the estimates downward, resulting in an underestimated egg mass density. To prevent this kind of bias, egg mass survey plot locations should be selected as if the sampler had no knowledge of egg mass distribution.

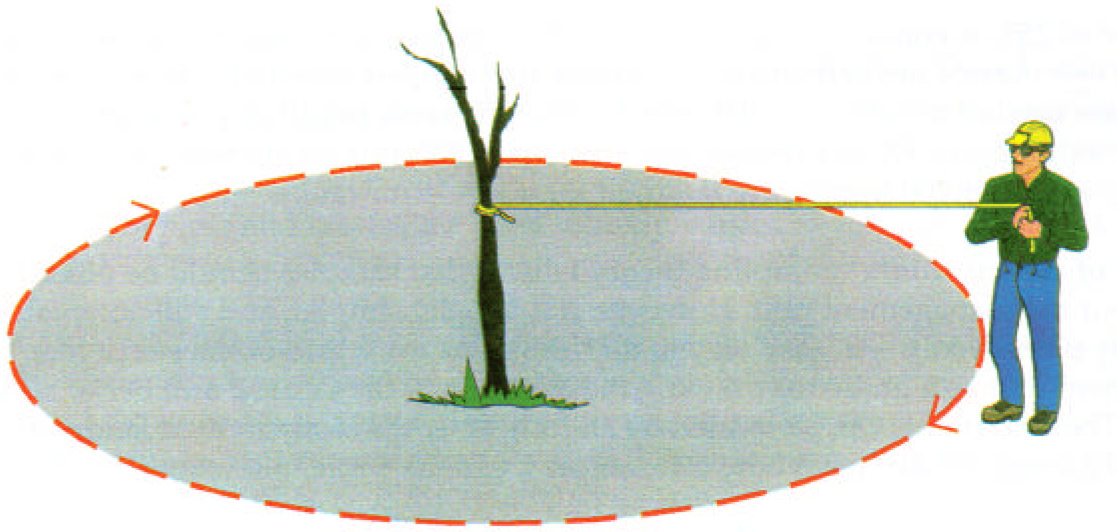

Fig. 3. Use of a rope for determining plot boundaries.

Plot layout. A circular plot is the simplest configuration to lay out. It is relatively easy to measure the perimeter of a circle by anchoring one end of a piece of rope or tape cut to a length of 18.6 feet (the radius of a 1/40th acre circle) and pulling the other end out to mark the perimeter (Fig. 3).

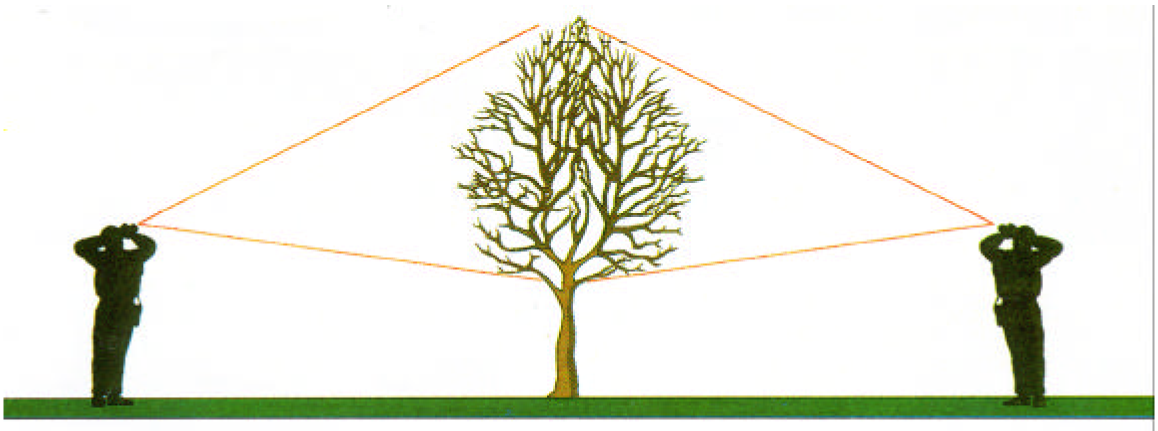

Fig. 4. Examination of tree crowns from two opposing vantage points.

Counting egg masses. The next step is to attempt to count all of the egg masses present within the boundary of the survey plot. First, count all egg masses located on trees with centers that fall within the perimeter of the plot. Binoculars must be used to see into the canopy. It will be necessary to examine each tree from different vantage points so that all sides of it can be seen. Small to medium trees will usually require examination from two opposed vantage points (Fig. 4). Large trees may require examination from more than two vantage points. Care should be taken to avoid counting the same egg mass more than once. This can be accomplished by scanning the trunk and branches systematically (e.g., top to bottom) with the binoculars. All objects on the ground, such as fallen branches and rocks, should be examined for egg masses (Fig. 5). This may involve moving these objects so that all surfaces can be seen. Weather conditions influence both the accuracy and speed of counting egg masses. Try, as much as is practical, to count egg masses under good weather conditions, which is when the sky is clear and the sun is overhead. Try to avoid poor conditions, which include dim lighting, long shadows (with the sun below a 45° angle), precipitation, and wet bark. It is virtually impossible to count egg masses on objects on the ground in the presence of a snow cover. Recent studies have shown that counts are more accurate after trees have dropped their leaves in the fall. However, in operational programs it is often not possible to wait that long to begin collecting egg mass count data.


A.


B.


C.


D.


E.


F.

Figure 5. *Typical locations for gypsy moth egg masses.*
A. Man-made objects, such as tires; B. At the base of tree trunks; C. Under bark crevices on tree trunks; D. On the underside at the base of limbs; E. On large rocks; F. Fallen limbs on the forest floor.

Table 1. Characteristics useful for differentiating new vs. old egg masses

Old Egg Masses	New Egg Masses
soft to touch	firm to touch
usually dull or bleached coloration	usually darker beige
exit holes present	no holes or small parasitoid exit holes present

Old vs. new egg masses. Egg masses that have recently been deposited usually differ in appearance from old egg masses that remain from previous seasons (Fig. 6, Table 1). New egg masses are usually darker in color and appear less ragged. It is generally not recommended to attempt to distinguish new and old egg masses visually. Therefore when making counts in the crown, count all egg masses, regardless of whether they appear old or new, and even if they are partially missing. A much more reliable determination of whether an egg mass is new or old can be made by touching it. New egg masses feel hard and full, while old egg masses feel soft and spongy. Of course, only egg masses within reach can be examined in this way. Therefore, examine by touch all egg masses that are within reach from the ground and determine the proportion of those egg masses that are new. This proportion can then be applied to adjust the number of egg masses counted in the crown. Because some survey plots may not have many egg masses available at ground level, data from all of the survey plots within a stand should be pooled together for the purposes of calculating the proportion of new egg masses. See Table 2 for an example of these computations.

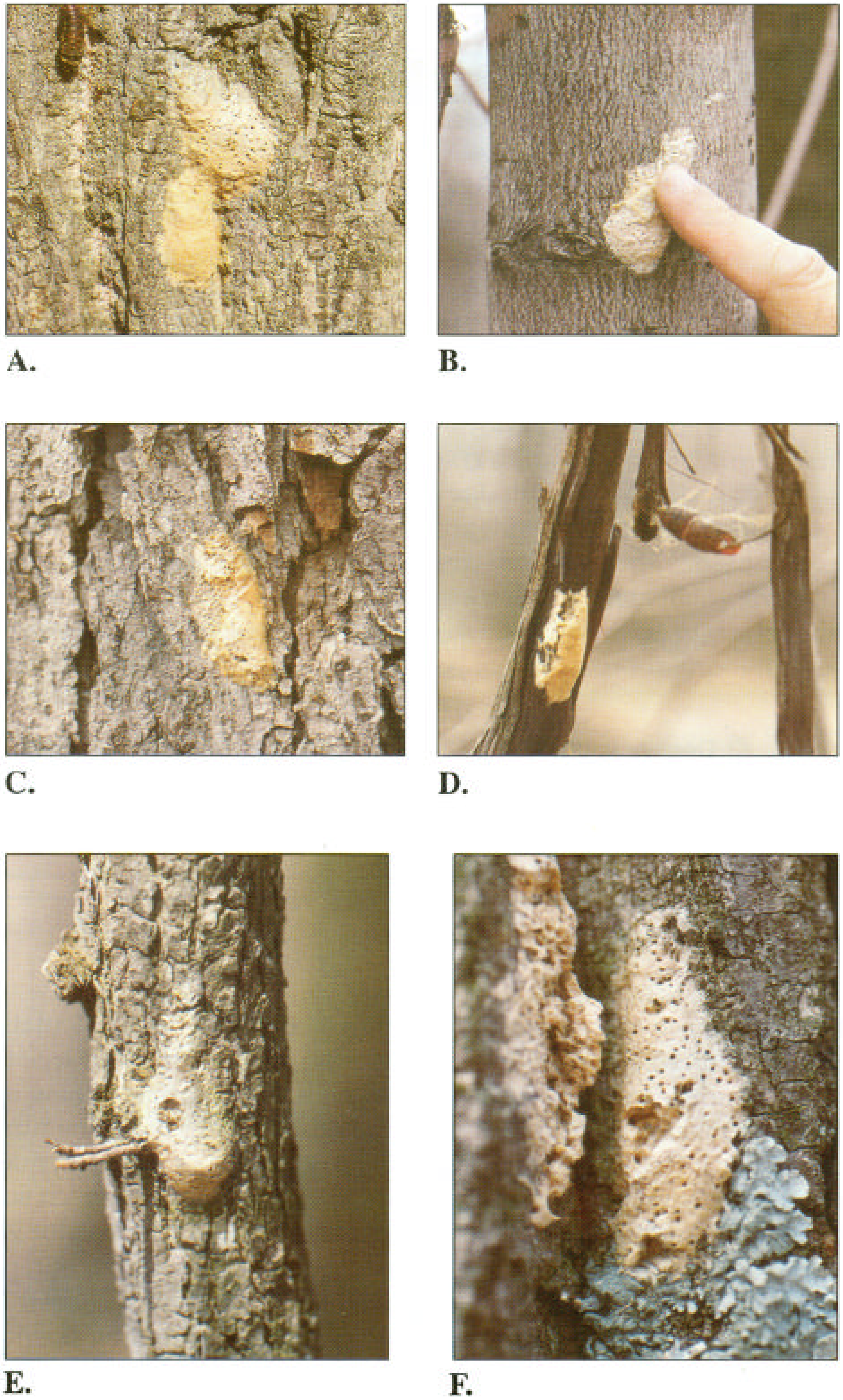

Figure 6. *Appearance of new vs. old egg masses.* **A.** *New and old egg mass;* ***B-D.*** *New egg masses;* ***E-F.*** *Old egg masses.*

Table 2. Sample calculation of egg mass density.

	Egg Mass Counts			Density
	Objects on Ground and Lower Portions of Trunks		Crown	(egg masses per acre)
Plot	**Old**	**New**		
1	2	7	2	**325**
2	6	3	0	120
3	3	7	6	414
4	3	2	4	170
5	8	9	14	674
6	1	0	3	67
7	0	1	2	85
❶ Totals	23	29		❹ mean = 221 ❺ SE = 90

❷ Proportion New = Total New / (Total New + Total Old)
= 29 / (29 + 23)
= 0.56

❸ Density in plot 1 = 40 (New + (Proportion New x crown))
= 40 (7 + (0.56 x 2))
= 325 egg masses per acre

Estimating egg mass density. The first step (1) in these calculations is to sum the total number of new and old egg masses located on the ground or lower tree trunks (i.e., within reach). These counts can then be used to estimate a proportion of new egg masses (2). The number of new egg masses in each plot is estimated by multiplying the number of egg masses that were out of reach (crown) by the estimated proportion of new egg masses and then adding the number of new egg masses that were within reach. Since each survey plot is 1/40th of an acre in area, the number of egg masses per acre at each survey plot is estimated by multiplying the estimated number of new egg masses in the plot by 40 (3). Step 4 is then repeated to estimate egg mass density for each of the plots. Finally, the mean density for the entire block is estimated as the average (sum of all plot densities divided by the number of plots) of plot estimates (4). It is important to also calculate the standard error (SE) of the estimated mean egg mass density(5), which can be used to determine how much confidence can be placed in the estimate. Means and standard errors can be calculated on some hand calculators, but it is best to enter the estimated egg mass density for each survey plot into a computer spreadsheet and request that means and standard errors be calculated. A useful way to establish the level of confidence that you can place in the estimated mean is to calculate a confidence interval, which indicates the probable range of values for the true mean. For example, the 80% confidence interval is the range of values within which one can be 80% certain that the true mean occurs. The following formula is used to calculate an 80% confidence interval:

$$CI = \text{mean} \pm (1.4 \times SE).$$

Thus, using the data given in table 1, the confidence interval for egg mass density would be:

$$221 \pm (1.4 \times 90) = 221 \pm 126.$$

Thus, we are 80% certain that the true egg mass density in the block is between 95 and 347 egg masses per acre.

Interpretation of survey results. There is no magic formula for interpreting the results of an egg mass survey. If the treatment threshold (the estimated egg mass density at which treatment should occur) is above the upper limit of the 80% confidence interval, then you can be 80% certain that the threshold has not been exceeded. Conversely, if the threshold is below the lower limit of the 80% confidence interval, then you can be 80% certain that the threshold has been exceeded. If the threshold falls within the 80% confidence interval, then it is likely that egg mass densities are near the threshold. However, the ultimate decision of what management actions should be taken must be made after consideration of available funds and manpower, sensitivity of host resources, and likely consequences of defoliation.

Prediction of Defoliation from measurements

Pretreatment egg mass density is related to subsequent defoliation (Fig. 7).

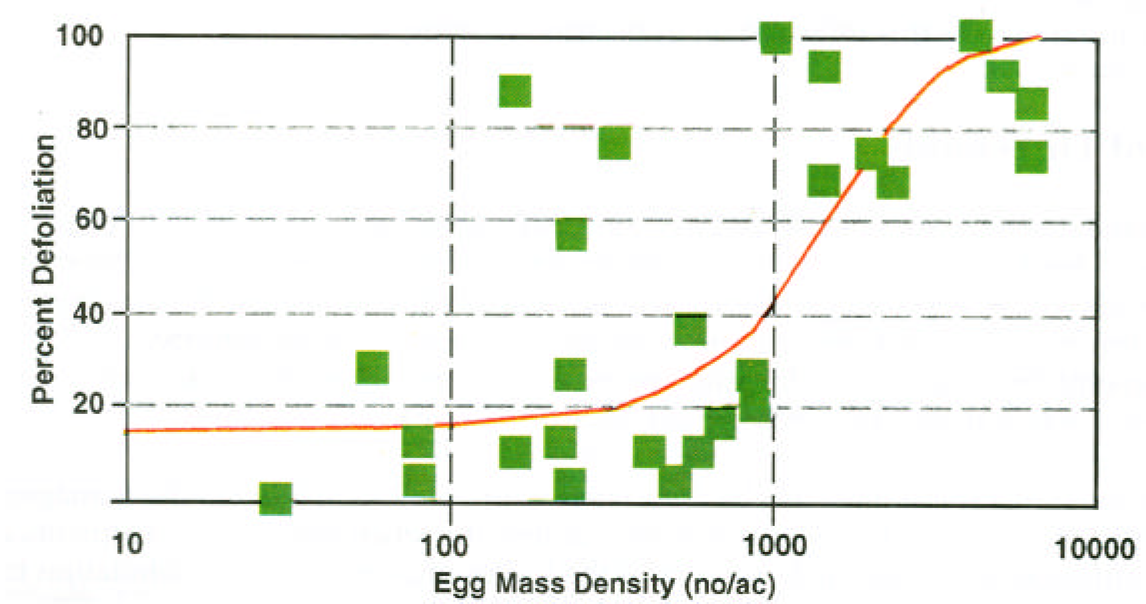

Fig. 7. Relationship between egg mass density and defoliation at several locations.

The squares represent data from stands where egg mass density and defoliation were measured. The solid curve is a statistically developed relationship. This line can be used for predicting defoliation by simply locating the estimated egg mass density on the horizontal axis, moving up vertically to the curve, and then following to the left to read the predicted defoliation level off of the vertical axis. One thing that is obvious from the wide scatter of observed points about this line is that there is a great deal of uncertainty in the relationship. Part of this uncertainty is due to measurement error in estimating density and defoliation, but the other uncertainty component is that the underlying relationship between "true" egg mass density and "true" defoliation is variable. This uncertainty is important to consider when making management decisions. The problem is particularly acute between densities of 100 to 1000 egg masses per acre where defoliation may vary between 0 and 100%, regardless of estimated density. In these situations, the manager must realize that this prediction of defoliation may not be correct and some attempt should be made to determine if the population is increasing or declining. When the management area is at the edge of the expanding front of the area generally infested by the gypsy moth, it can often be assumed that the population is increasing. However, if the area has been infested for several years, other information will be necessary to make this determination.

Egg mass size can provide some information about the condition of the gypsy moth population. Small egg masses (i.e., < 20 mm in length) are indicative of a declining population. Large egg masses (i.e., > 30 mm in length) are indicative of an increasing population.

The proportion of old egg masses can also be useful in characterizing the population condition. Large numbers of old egg masses (i.e., > 50% old) is indicative of a declining population, whereas a very low proportion of old egg masses (i.e., < 25% old) suggests that the population is building.

Another factor that might enter into the decision making process at this point would be the regional conditions. If egg mass sampling indicates a low to moderate density population in a stand, the presence of high density populations in surrounding areas will likely increase the probability of defoliation. Similarly, a moderate to high density population surrounded by low density or declining populations is probably less likely to cause defoliation. Therefore, the prudent manager should consider population levels and trends in the region surrounding the area of interest and integrate this information in decision-making.

Management Thresholds

Before conducting an egg mass survey, land managers should have clearly defined management objectives. These objectives will help in determining areas for survey (management unit), the intensity of the survey (desired precision), and guidelines for establishing intervention thresholds (treatment). For most gypsy moth programs, the management objectives are either nuisance abatement, prevention of defoliation, prevention of tree mortality, or a combination of these objectives. Each of these objectives should have its own intervention threshold.

Information on the impacts (damage levels) associated with defoliation will aid the manager in establishing thresholds. These thresholds will help determine when and where treatments are needed. In a hardwood forest, about 30 percent of the leaves must be eaten for defoliation to become noticeable from the air. Studies have shown that growth loss begins at about 40% defoliation, and is proportional to the percent of defoliation thereafter. Refoliation occurs when about 60% of the foliage is lost. Refoliation results in the loss of a tree's stored reserves, which can cause growth loss and a reduction in the tree’s overall health and survival.

The density of gypsy moths that can result in concern or nuisance in residential or recreational settings has not been well established. For some individuals the sight of a few larvae may be intolerable, while others may not be concerned until defoliation is evident. Unsuspecting individuals may not even notice defoliating populations.

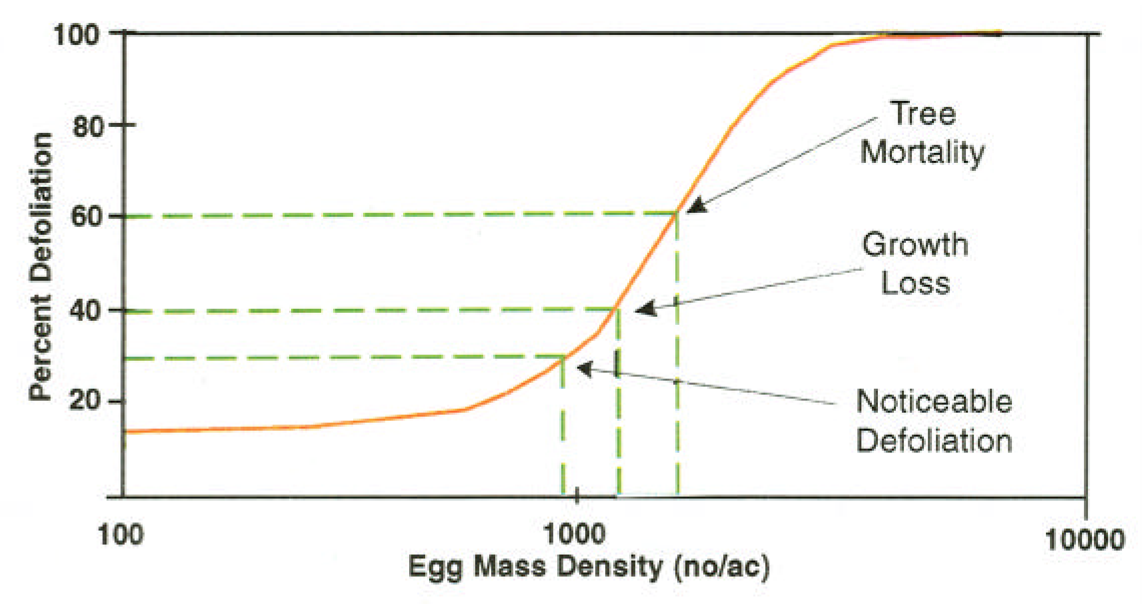

Fig. 8. Relationship between defoliation and egg mass density thresholds for three damage criteria.

Selection of thresholds by management objectives is demonstrated in Figure 8. This figure shows the same defoliation prediction curve that was illustrated in Figure 7. For the prevention of noticeable defoliation (>30%) an intervention threshold value of 500 to 750 egg masses per acre might be appropriate. To prevent growth loss (> 40% defoliation) then 700 to 900 egg masses per acre could be set as a threshold. Finally, to prevent mortality a threshold of 1000 to 1400 could be used. The manager may chose to modify these to lower levels if the stand has been subjected to other stresses that may predispose trees to mortality or if unusually high-value or specimen trees are involved. Experience in survey and knowledge of local forest condition will help establish threshold values that meet your management objectives.

An intervention threshold value of 250 egg masses per acre has been widely used in the past for intervention in both general forest and residential areas. While this value may be justified for reducing certain nuisance impacts, it may not be justified for other management objectives. If a manager's objective is to prevent noticeable defoliation, growth loss, or mortality, then initiating treatment at 250 egg masses per acre would show little or no return on the expense of treatment. Of course, Fig. 7 illustrates that there is considerable uncertainty about the relationship between egg mass density and defoliation. Therefore, the significance of defoliation risk to management objectives should be a consideration when establishing intervention thresholds. While selection of an intervention threshold of 250 egg masses per acre may result in the needless treatment of many stands that would never become defoliated, it does serve to minimize any risk of >30% defoliation.

Caveats and Concerns

The procedures described here represent a scientifically-based approach to decision-making for gypsy moth managers. Unfortunately, these procedures are far from perfect; it is impossible for managers to predict defoliation levels without a certain amount of error. These errors come about because of 1) the high cost of egg mass surveys often leads to inadequate replication, which may result in considerable error in measuring egg mass densities and 2) there is considerable variation in the relationship between actual egg mass densities and subsequent defoliation.

A technique known as "sequential sampling" has been developed to help managers deal with the first problem mentioned above[6]. Under this procedure, the variation in counts among fixed radius plots within a management unit are used to advise managers whether it is necessary to continue sampling at more plots in order to classify an egg mass population as being either above or below an action threshold given a

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[6]Fleisher, S, J. Carter, R. Reardon, and F. W. Ravlin. “Sequential Sampling Plans for Estimating Gypsy Moth Egg Mass Density” AIPM Technology Transfer NA-TP-07--92.

fixed error level. While this method may often be helpful in guiding the collection of egg mass data, the problem remains that it is often very expensive to collect enough egg mass samples to make a sound treatment decision.

Figure 7 illustrates the uncertainty in predicting defoliation when egg mass densities range from 100 to 1,000 egg masses per acre. Following the guidelines illustrated above should be useful for guiding treatment decisions for populations in these situations but there will continue to be errors in the decision-making process. Research currently underway may eventually improve this situation. The USDA Forest Service, Forest Pest Management field offices listed below can be contacted to obtain the latest information on egg mass sampling procedures:

Forest Health Protection
USDA Forest Service
180 Canfield St.
Morgantown, WV 26505
(304) 285-1540

Forest Health Protection
USDA Forest Service
PO Box 640
Durham, NH 03824-9799
(603) 868-7704

Forest Health
USDA Forest Service
200 Weaver Blvd.
Asheville, NC 28802
(704) 257-4321

Forest Health Protection
USDA Forest Service
1992 Folwell Ave.
St. Paul, MN 55108
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