Appendix E - Entomological Work Plan

Sampling Objectives

- Verify that Douglas-fir tussock moth (DFTM) populations are at high suboutbreak or higher levels in areas to be treated (10+ larvae/1000 sq. inches of foliage, midcrown).
- Ensure proper timing of insecticide application.
- Estimate pre-treatment population densities of DFTM.
- Estimate post-treatment population densities (28 days, 35 days after treatment) of DFTM.
- Determine whether specific project objectives were accomplished relating to protection of foliage.

Definitions

In this document, terms are used in reference to an operational suppression project that may be unfamiliar to the reader. To avoid confusion or misunderstanding we will define several of the terms used in this Chapter or in other places of the Operation Plan. (In addition, a full listing of literature citations can be found in Appendix H.)

Analysis Area - a geographic area delineated on a map that encompasses both insect host-type and non-host areas comprising one or more high-value resources that are linked to, or influenced by, stands of conifer trees which are at risk of being directly damaged or killed by insect defoliation, or may be damaged or killed by subsequent infestations of secondary insects or diseases, or by wildfire as a result of insect-caused woody fuel accumulations. Analysis Area is the primary area upon which an environmental analysis is conducted when a proposed federal action is being considered to manage insect populations on a Ranger District, National Forest, or several National Forests within a Region.

Analysis Unit (AU) - a geographic area derived from the Analysis Area that is delineated on a map and encompasses primarily the specific high-value, high-risk, host-type stands that may be treated in an operational insect suppression project. In addition, it may contain some inclusions of lower value, lower risk, host-type stands, as well as non-host areas.

Host Acres - Pure stands, or mixed stands composed of at least 20 percent or more of the following Douglas-fir tussock moth susceptible tree species; Douglas-fir, grand fir, white fir, and subalpine fir.

Host Type - The preferred vegetation of the Douglas-fir tussock moth. In the case of this insect, the preferred species of tree is Douglas-fir, grand fir, white fir, and subalpine fir (modified after USDA 2000).

Lower Crown Sampling - Method of estimating an index of population density by the nondestructive sampling of 18-inch branches in the lower crowns of trees (Brookes, et al. 1978).

Midcrown Density - A measure of insect population density estimated for, or obtained directly from, that portion of the tree with main branches originating from the middle one-third of the bole (Brookes, et al. 1978).

Nucleopolyhedrosis (NP) - Viral disease of insects, mainly larvae of certain Lepidoptera and Hymenoptera, characterized by the formation of polyhedra in the nuclei of infected cells; usually fatal (Brookes, et al. 1978).

Operational Control Project - Project conducted to control a forest pest, using information accumulated from a sequence of tests Blaboratory bioassays, field experiments, and pilot control projects (Brookes, et al. 1978).

Outbreak (tussock moth) - Period of high insect numbers, during which conspicuous defoliation occurs; characterized by four phases Brapid increase (release, phase I), peak (II), decline (III), and postdecline (IV) (Brookes, et al., 1978).

Outbreak Population - Larvae are abundant, causing defoliation that is usually visible in aerial or ground reconnaissance; density of larvae is greater than 20 first and second instars per 1,000 in^2 of midcrown branch area (Brookes, et at. 1978).

Spray Block - an area delineated within an Analysis Unit based on a stratification of the topographic characteristics (usually within a specified elevation band, aspect, slope, and possibly other factors) that might influence the operational characteristics or production capabilities of the application aircraft, and that might influence the phenology (development), abundance, and distribution of the target insect over the treatment area. Spray Blocks are usually drawn on a USGS Quad Map in such a way to make them as uniform in their defining characteristics as possible. This typically encompasses only the acres that can be safely and reasonably treated within the daily operating window and operating characteristics and limitations of the application aircraft.

Suboutbreak Population - Larvae are common but have not reached outbreak numbers; density is between 2 and 20 first and second instars per 1,000 in^2 of midcrown branch area; has the potential to increase to an outbreak population (Brookes, et at. 1978).

Treatment Acres - This refers to the acres within an Analysis Unit that would receive an application of insecticide for biological or logistical reasons.

Sampling Design

Cocoon density plots, development-monitoring plots, and pre- and post-treatment evaluation plots will be established in each accessible Analysis Unit (AU) to accomplish the above objectives. Cocoon density plots will be established with a minimum of one plot per 1,000 acres or 50 Plots per AU, whichever is higher. A partial Analysis Unit will require a minimum of 25 cocoon density plots before it can be dropped from consideration from treatment due to low population numbers. Before an entire Analysis Unit can be dropped from treatment due to low populations, a sample total of at least 50 cocoon density plots is required. Each Analysis Unit will be stratified by spray blocks. They will be based on factors such as elevation, terrain, slope and aspect, operational characteristics, and probable insect phenology, abundance, and distribution characteristics. Each accessible spray block will contain three 10-tree development monitoring plots. Each accessible spray block will have a minimum of one 5-tree pre- and post-treatment evaluation plot, with a minimum of 50 such plots per Analysis Unit. For each individual Analysis Unit, one untreated control area similar in character to the AU will also be established with 50 five-tree pre- and post-treatment evaluation plots. From this, comparisons can be made between treated and untreated areas.

Qualification of Analysis Units for Treatment

Cocoon densities will be used to initially qualify Analysis Units (AU) for treatment. The procedure outlined by Mason et al. (1993) will be followed to assess cocoon densities over each AU in a one-time procedure to qualify or modify the boundaries of each AU for potential treatment. Establishment of fifty, 50-tree cocoon density plots per Analysis Unit will allow prediction of mean larval densities over any individual AU with a precision adequate for operational treatment purposes (a standard error equal to or less than 30 percent of the mean. Mason et al., 1993). If more than 50 cocoon density plots are established over an Analysis Unit, the sampling precision may approach ten percent or less of the mean for the whole AU. This sample will be made only once for each AU. It should be timed to occur as early as possible, but can be done anytime **before** egg mass hatch.

To estimate a mean population density from lower crown cocoon samples that predicts a treatable high suboutbreak (10 larvae/1000sq. in. foliage) or higher population of tussock moth (i.e., 3.75 cocoons per 50 trees or higher), crews will conduct cocoon sampling on 50 or more (see above) plots. These will consist of 50 Douglas-fir tussock moth host trees (Douglas-fir, true firs) scattered over the AU to represent varying conditions of topography, elevation, aspect, etc., on the AU. Host trees for this sample can be any size larger than

approximately 20 feet tall. Crews will examine the underside of three 18-inch branch tips from each of the 50 trees per plot. The number of new (1999) cocoons and egg masses will be recorded on the Douglas-fir Tussock Moth Cocoon Sampling Form (see Appendix A), and these data summarized on BIO. FORM NO. 1. Be sure that all copies of the completed Douglas-fir Tussock Moth Cocoon Sampling Form for the plot are stapled on the summary form or BIO. FORM NO. 1 (refer to Appendix B - for all spray operations forms).

If after sampling cocoons, a portion of the Analysis Unit seems to have low numbers of cocoons present on host trees, then additional plots should be added to this portion of the AU to validate these low cocoon densities. This information may be used by the Project Entomologist to split an AU where one portion may have DFTM populations high enough to warrant treatment, but the other half doesn't.

Using the cocoon density data, the Project Entomologist will review the initial stratification of the Analysis Unit into spray blocks and may redefine the boundaries of the Spray Blocks or Analysis Units as required. If after installing additional cocoon density plots, the numbers are close to the treatable population threshold, the Project Entomologist may postpone his/her decision to drop spray blocks in that portion of the Analysis Unit until the pre-spray population sample is taken. Remember, it is the **Pre-spray Larval Density sample**, NOT the cocoon density sample, that ultimately determines if an adequate population exists on a spray block to warrant treatment based on the treatment threshold.

Development Plots Selection

Development sampling will require the establishment of three 10-tree plots in each spray block accessible by project vehicle, or within a relatively short hike (one mile) from a road if the block is less accessible. Flag lines should be used in addition to written directions and a map drawing to facilitate easy location of more remote plots by sampling crews on spray blocks with poor or no vehicle access. Plot sites will be in stands with predominately host species (Douglas-fir or grand fir) and with trees which have newly developing buds on branches that can be reached from the ground. Each plot should have at least five additional host trees (at least 20 feet tall) which are not overtopped by other DFTM host trees and have branches with newly developing buds that can be reached with lower crown sampling equipment. This will allow establishment of an Evaluation Plot at the same site if necessary. A map of the Development Plot location will be drawn on the bottom or back of BIO. FORM NO. 2 (refer to Appendix B for all spray operations forms).

Egg Mass Tagging and Monitoring

At each Development Plot site up to ten new DFTM egg masses will be tagged (on the tag record the following: Block No., Plot No., Egg Mass No.) and flagged with a flagging color combination approved for the Project on the ten development plot trees so they can be monitored. No more than two egg masses will be tagged on any individual tree, and no more than ten at the plot site. Only egg masses that are easily within reach from the ground should be tagged. These egg masses can be on trees of any size and of either host species. If this is also the site of an Evaluation Plot, NO EGG MASSES ARE TO BE TAGGED ON EVALUATION PLOT TREES.

When DFTM egg masses are being located and tagged at each of the development plots, if they are "easily found" this will be "circumstantial" evidence that the populations are at outbreak levels and no larval density estimates will be required before the PRE-TREATMENT DENSITY SAMPLE. If a crew of two has not located at least three egg masses within 15 minutes, or a person working alone has not located at least three egg masses in 30 minutes, they should proceed on to the next plot site. This will indicate that the populations are relatively low and that larval density samples may need to be collected to assure that the DFTM populations are at suboutbreak or higher levels (refer to the LARVAL DENSITY SAMPLING section for direction on what to do if this occurs). It may be necessary to flag a compass line in to each sample tree to help crews find each tree with an egg mass during subsequent visits to measure egg mass hatch and larval development (**be sure to indicate these flag lines on the plot location map**

drawn on BIO. FORM NO. 2. Refer to Appendix B.)

Crews will monitor all ten of the flagged egg masses on each plot in two-day intervals until the neonate (newly emerged) larvae have completely dispersed from the egg mass and onto the foliage. The plots will be revisited every other day to monitor egg masses for appearance of newly hatched larvae. When each egg mass begins to hatch (first-instar larvae appear and rest atop the egg mass), the date that hatching first occurs, or was observed, will be recorded for each egg mass on each of the plots. In addition, the condition of foliage development on the tree containing the egg mass or egg masses being monitored will be recorded at each visit to the plot. Foliage development will be recorded as one of the following conditions: (1) Bud caps tight; (2) Bud caps swollen; (3) Budburst with foliage unfurling; or (4) Bud caps dropped with new needles fully exposed. All egg mass and foliage development data will be recorded at the time of each visit on BIO. FORM NO. 3 (refer to Appendix B for all spray operations forms)

In addition, 2 live larva will be collected per plot. They will be individually placed into small tight-lid plastic petri dishes, (Falcon No. 1006, one larva per dish) and labeled with the A.U. #, spray block #, Plot #, date and collector's initials. This will be a one-time collection at the time of first egg hatch from each of the egg mass sites. A small sprig of new DFTM host foliage will be placed in the dish with the larva to provide food while the larva is transported back to the lab at Project Headquarters. BE CAREFUL NOT TO SMASH THE LARVA BETWEEN THE FOLIAGE AND THE PETRI DISH LID OR BOTTOM. Dishes containing live larvae or live insects should be placed in a cooler containing Blue Ice to keep them cool until they can be delivered to the lab. Petri dishes **SHOULD NOT** be placed in direct sunlight or left in a closed vehicle without a means of refrigerating the specimens or providing other means of keeping them cool. The larvae collected in petri dishes will then be transported to the Forestry and Range Sciences Laboratory in La Grande, Oregon. There they will be reared on artificial media until death or pupation for determinations of parasitism and incidence of naturally-occurring egg mass Nucleopolyhedrosis Virus (NPV).

Larval Development Sampling

Development plots will be established in both treated and untreated control areas. The larval development sampling will commence at the time when complete larval dispersal is first observed. At each development plot, larval development samples will be collected from three lower crown 18-inch branch tips from each of ten trees per plot using lower crown beating techniques. AFTER EACH BRANCH TIP IS SAMPLED, THE TIP WILL BE REMOVED USING A HAND CLIPPER (this is to avoid sampling a previously sampled branch tip in subsequent visits to the plot). These trees can be any height greater than 20 feet and should be the same species as the predominate host tree at the plot site. DFTM larvae will be classified by instar in the field (see Beckwith, 1978). If the collectors are not confident of their ability to determine instars in the field, then all larvae from each tree (i.e., all larvae from the three branch tips) will be placed into a separate container, labeled, and brought back to the laboratory for determination by the Entomologist. All larval development data will be recorded on the Larval Development Data Entry Sheet (BIO. FORM NO. 4--Refer to Appendix B).

At one of the Development Monitoring Plots and/or a minumum of 50 sites per A.U. an additional five host trees will be flagged and tagged and used for the pre- and post-treatment evaluation sampling. These trees on the pre- and post-treatment evaluation plots will remain undisturbed until after larvae have completely dispersed from the egg masses based on the monitoring of the Development sampling trees on the plot with flagged egg masses, and the block is deemed ready to release for treatment.

Larval Density Sampling (This procedure is to be used only if egg masses are hard to find)

If egg masses are difficult to locate in several contiguous spray blocks, the Project Entomologist or Assistant Entomologist will decide whether to initiate larval density

sampling in these areas. If initiated, a lower crown sequential sampling method (Mason, 1978 and 1979) will be used to determine if populations are at least at high suboutbreak levels (i.e., 10 larvae/1000 sq. in. foliage area, midcrown). Sampling will not commence until larvae have dispersed from the egg masses onto new foliage. This should coincide with the time when all buds on host trees have burst and average growth of new shoots has reached one to two inches (Mason 1978).

At each established plot site and at enough other sites within the area of interest to make a total of at least 30 plots, three lower crown 18-inch branch tips will be sampled until a decision is made on classification of the population (low or suboutbreak) or 20 trees have been sampled, whichever comes first (see BIO. FORM NO. 5 [refer to Appendix B for all spray operations forms]).

The following instructions for conducting sequential sampling of tussock moth populations in the lower crown are taken verbatim from Mason (1979):

1) Randomly select a host tree (**other than a tagged evaluation plot tree**) for sampling that has foliage within reach from the ground. A tree of any height is acceptable as long as sample branches have new growth. Avoid sampling the tops of small trees.

2) Hold a drop cloth under 18-20 inches of the branch to be sampled. Rap the branch vigorously with a short stick, being careful not to disturb other branches on the tree. Larvae will drop onto the cloth.

3) Examine the drop cloth for tussock moth larvae. If a larva is found, note the tree as infested and proceed to another tree. If no larva is found, continue sampling other branches on the same tree until a larva is found or until a total of three branches have been examined.

4) Record the number of infested trees on the sequential form (BIO. FORM NO, 5 - Refer to Appendix B) by adding each one to the total number of infested trees found previously. To record a tree as uninfested, add zero to the previous number and write the figure under the appropriate number of sample trees.

5) If the cumulative total of infested trees is equal to or less than the low value in the sequential sampling plan on the form, or equal to or more than the high value, stop sampling and classify larval density as either low or suboutbreak, respectively. If the cumulative total is between the specified limits in the overhead box, no classification can be made and another tree must be sampled.

6) If larval density is not classified after 20 trees have been sampled, stop sampling and classify the plot in the intermediate class.

Classifications of all plots within the area of interest will then be taken into consideration by the Project Entomologist or an Assistant Entomologist in deciding whether to exclude that particular area from insecticide treatment.

Aside from determining whether or not these borderline areas should be treated, early larval lower crown density samples, coupled with the larval development samples on individual spray blocks will help support the timely data needs for the air operations group to estimate treatment production characteristics and aircraft support requirements based on application aircraft production capabilities before the project treatment actually begins.

Releasing Spray Blocks For Treatment

The primary development sampling data will be used to time the release of the spray blocks for treatment. Blocks will be released when 60 percent of the DFTM larvae collected in a spray block are in the second instar (L2). The release data will be documented on BIO. FORM NO. 6 (refer to Appendix B) by the Project Entomologist or an Assistant Entomologist. If a spray block has very poor access, and no plots are established in it, it will be released for treatment at the same time as the nearest block

with similar elevation and aspect. This will be at the discretion of the Project Entomologist or an Assistant Entomologist.

At this target age (60 percent L2), we anticipate the distribution of instars will also include some third instars (and possibly a minor number of fourths), as well as some first instars. The release date at which this distribution occurs is estimated to be approximately 10 days (plus or minus a few days) after egg hatch. This release timing allows for the treatment of an insect stage that will result in the maximum infection rates and secondary passages of the virus, based on studies by Stelzer et al. (1977). The direct mortality of tussock moth larvae from application of virus may occur within 12 to 14 days after application, and the second passage of infection (second wave of virus) has been noted to occur 21 and 35 days after application (Stelzer et al. 1975).

Treatment of the population must occur within 72 hours of release, or the population will have to be resampled to determine the current pre-treatment densities. If the spray block has not been treated within 12 to 14 days of release, the Project Entomologist will consider withdrawing that block from treatment, since the second wave of virus infection that will affect the larvae surviving the initial application may not occur before the late-instar (L5 and L6) larvae begin to pupate, if the application is delayed more than 12 or 14 days. There may be some occurrence of pupal-stage virus-caused mortality in pupae that were infected in the late instars; however, because the virus mainly infects the larval stage this amount of pupal-stage virus-caused mortality is probably negligible in terms of the overall course of the disease epizootic. A greater degree of population reduction, and especially greater foliage protection, will occur from applications of nucleopolyhedrosis virus when the larvae are initially infected during earlier instars. On this infection schedule, the majority of larvae would be assured of dying from nucleopolyhedrosis (NP) in both the first and second waves of the virus, before reaching pupation.

Evaluation Sampling Design

In addition to the Development Monitoring plots established to monitor development in each accesssible spray block, crews will establish a minimum of fifty Evaluation plots in each treated and in untreated control areas. For each Evaluation plot, an Evaluation Plot Location Data Sheet (BIO. FORM NO. 7 - refer to Appendix B) will be prepared by sampling crews. Larval populations within these areas will be sampled in order to compare post-treatment densities with pre-treatment densities, and compare populations on treated blocks with those on untreated blocks.

Crews will establish a minimum of one treatment evaluation plot within each accessible spray block, and 50 other evaluation plots in one untreated control area for each AU with a minimum of 50 plots per analysis unit. The untreated control area must contain at least suboutbreak populations (10 larvae/1000 sq. in of foliage, mid crown) of tussock moth.

In each evaluation plot, five medium-sized trees (20 to 45 feet tall) which are not overtopped by other DFTM host trees and have branches with new foliage that can be reached from the ground will be flagged (with a flagging color combination approved for the Project) and labeled (with a white tag with AU No., Block No., Plot No., and Tree No.). Trees of the predominate species (Douglas-fir or grand fir) present at the plot site will be selected. These trees will be the ones from which pre- and post-treatment density samples will be collected. **No other samples are to be collected from these trees except for collections of live larvae for rearing to determine NPV infection levels (see below).**

Pre-treatment Larval Sampling

When a spray block has been released for treatment, pre-treatment larval samples will be collected as soon as possible from the designated, flagged, pre- and post-treatment evaluation trees.

Crews will sample these trees by beating three 18-inch lower crown branch tips from each of the five sample trees per plot using the lower crown beating technique. The numbers of DFTM larvae observed on the beating cloth

from the three branch tips will be recorded by instar on BIO. FORM NO. 8 (refer to Appendix B). AFTER EACH BRANCH TIP IS SAMPLED, THE TIP WILL BE REMOVED USING A HAND CLIPPER (This is to avoid sampling a previously sampled branch tip in subsequent visits to the plot). If possible, larval instar determinations will be made in the field and recorded on a data form. If this is not done, larvae collected from all three branches on each tree will be put into a labeled vial of alcohol and brought back to the laboratory to be checked by the Project Entomologist.

Likewise, a pre-treatment sample will be obtained, and larvae collected from all plot trees on the untreated control block(s). Data will be recorded on BIO. FORM NO. 8 (refer to Appendix B) and clearly indicated that sample data is from a pre-treatment-timed Control Block sample by recording the words: "**UNTREATED CONTROL BLOCK**" at the top of the data form.

If a block is not treated on or before the third morning after the block has been released for treatment, another pre-treatment sample will be collected. This will be done to provide a reasonable estimate of larval instar distribution at the time of treatment. This data may be important in explaining the results of the treatment with the virus, especially if mortality is lower than expected and can be explained by instar distribution at the time of treatment.

Pre-treatment Larval Collections

To determine pre-treatment levels of NPV and parasitism, crews will collect two live larvae from each sample tree on each plot (from both treated and untreated control blocks) and place them individually into small tight-lid plastic petri dishes (Falcon Petri Dish No. 1006; one larva per dish), and label the dish with the AU Number, Spray Block Number, Plot Number, date of collection, instar, and collector's initials. A small sprig of new DFTM host foliage will be placed in the dish with the larva to provide food while the larva is transported back to the lab or Project Headquarters. Dishes containing larvae should be placed in a small styrofoam cooler containing Blue Ice to keep them cool until they can be delivered to the lab.

Petri dishes with live insects should **not** be placed in direct sunlight or left in a closed vehicle without a means of refrigerating the specimens or providing other means of keeping them cool (such as with an ice chest containing Blue Ice or Party Ice). These larvae will be transferred to a clean petri dish containing a small piece of artificial insect diet at the lab, the dish marked with the same information as on the original dish, and the petri dishes containing larvae transported to the Forestry and Range Sciences Laboratory in La Grande where they will be reared on artificial media until death or pupation, for determinations of parasitization rates and incidence of naturally-occurring NPV.

Post-treatment NPV Infection Rate Sampling

In addition to sampling at 28 days and 35 days after treating to determine reductions in population densities, crews will make the last larval collections from the evaluation plot trees at the 7th to 10th day after treating each block. This sample will be used to determine Nucleopolyhedrosis (NP) prevalence rates resulting from the initial application of TM BioControl-1 in field-collected larvae that are reared in the laboratory. Crews will once again collect two live larvae from each sample tree on each plot (from treated and control blocks) and place them individually into large plastic petri dishes (Falcon Petri Dish No. 1029; one larva per dish), and label the dish with the AU Number, Spray Block Number, Plot Number, date of collection, instar, and collector's initials. A small sprig of new DFTM host foliage will be placed in the dish with the larva to provide food while the larva is transported back to the lab or Project Headquarters. Dishes containing larvae should be taped close with scotch tape or masking tape to prevent escape of larvae. The dishes of larvae will be placed in a small styrofoam cooler containing Blue Ice to keep them cool while returning to the lab. Petri dishes with live insects should **NOT** be placed in direct sunlight or left in a closed vehicle without a means of refrigerating the specimens or providing other means of keeping them cool (such as with an ice chest containing Blue Ice or Party Ice). These larvae will be transferred into a clean petri dish containing a small piece of artificial insect diet at the lab, the dish

marked with the same information as on the original dish, and the petri dishes containing larvae transported to the Forestry and Range Sciences Laboratory in La Grande where they will be reared on artificial media until death or pupation, to determine parasitism rates and incidence of naturally-occurring NPV. **Note: The same precautions must be taken again when collecting larvae from untreated and treated sample trees. The untreated control plots must be sampled <u>before</u> any of the treated plots to avoid contaminating the untreated larval samples. The beating cloths should be laundered in hot soapy water before beginning each control block sampling, at each sampling period. Forceps used to collect larvae in petri dishes must be kept in a soapy water between each larval collection to insure that NPV is not accidentally transmitted between samples.**

<u>Post-treatment Larval Sampling</u>

The Evaluation Plots will be re-visited, and the flagged and tagged sample host trees will be re-sampled at 28 days, and again at 35 days (plus or minus one day) after treatment. Samples will be obtained from **THE SAME** trees from which the Pre-treatment samples were collected. This will give data from which we can estimate percent population reduction and get a population density to help predict population levels for the coming year.

Both the treated spray blocks and the untreated control blocks will be sampled during these post-treatment timings. These post-treatment sampling periods will provide data from which we can estimate percent population reduction.

Crews will obtain samples by beating three 18-inch branch tips from each of the five trees per Evaluation Plot using the lower crown beating technique. **DO NOT SAMPLE BRANCH TIPS ADJACENT TO THOSE THAT WERE CLIPPED IN PREVIOUS SAMPLES.** The numbers of tussock moth larvae (and pupae if present) that fall onto the beating cloth will be recorded by instar on BIO. FORM NO. 9 (refer to Appendix B). If possible, larval instar determinations will be made in the field. If this is not done, larvae collected from all the branches on each tree per plot will be put into a labeled vial of alcohol and brought back to the laboratory. DFTM larval instar determinations will be made in the field.

35-Day Post-Treatment Defoliation Monitoring And Tree Damage Prediction

Population treatments to suppress tussock moth in areas of critical resource concern are intended to address the following project objectives (USDA 2000):

- Protect habitat for Threatened and Endangered species, specifically salmon, steelhead, bull trout, wildlife nesting habitat, designated old-growth, and late and old structural stands.
- Protect Health and Safety areas, including residential and administrative areas, high use developed recreation areas, municipal watersheds, and designated scenic areas.
- Protect High Investment areas, such as seed orchards, areas currently being protected from bark beetles, and sold timber sales.

Insecticide treatment is needed to protect and preserve vegetation necessary to maintain the desired habitats and other resources and amenities described above. Additional plots will be installed to measure defoliation amounts. These plots will be installed in each Analysis Unit, including Control units. 25 plots will be installed in each AU. Each plot will consist of 20 trees. These trees will be dominants and co-dominants of host species. These trees will be surveyed by eye, so no attention needs to be paid to finding trees with branches easily reached from the ground. Plots will be evenly spaced throughout the AU. The first tree on each plot is the plot center, tree number 2 is north of tree number 1 and then trees are numbered clockwise. Plots need to be located in areas which will be sprayed, except for plots within the control units. Each tree will be marked with paint with plot # and tree # with a painted band on the tree. No blue-marked trees will be chosen. Chosen trees need to be large enough to have numbers painted on their trunks. Trees within the plot will be grouped around the central location, or tree number 1. Plot location will be identified with a latitude and longitude. A map of the trees will be placed on the placed on the back of the form. A narrative concerning the plot will be done on the front of the form.

Table V-1: Annotated Table of Tree Defoliation Classes by Percent of Crown Totally Defoliated (reproduced from Wickman, 1979)

Defoliation Class	Percent Crown Defoliation	Description
0	0	No defoliation
1	10	Top ten percent of the crown (mostly new foliage) is totally defoliated. Additional defoliation of new foliage may occur lower in the crown, but branches are not completely stripped of needles.
2	25	Branches are completely stripped of needles in the top quarter of the crown. This crown area is mostly new foliage, but some feeding on older foliage may also occur at this level. Most new foliage is removed lower in the crown.
3	50	The top half of the crown is totally defoliated. There is significant feeding on older needles at this level of defoliation. All new foliage is damaged in the lower half of the crown.
4	75	The top three-quarters of the crown is totally defoliated. There is heavy feeding on older foliage and all new foliage is removed from the remainder of the crown. The crown may take on a very ragged or uneven appearance below the area of total defoliation.
5	90	Green needles remain on only the lowest 10 percent of the crown. Sometimes only the lower whorl of branches is left with older green needles.
6	99	The tree may have only a few green needles remaining on one of the lower

		branches.
7	100	Trees are completely stripped of foliage. There may be some stubs of green needles remaining, but these will eventually be lost. At this level of defoliation, even the new buds may be eaten.

Forecast of damage to the host trees on the spray blocks will be determined from the seven defoliation classes described above on the Evaluation Plot sampling trees from all plots on each spray block and untreated control block. These data will be summarized by treated Analysis Unit. The effect of defoliation on tree mortality will be forecasted using the criteria given by Wickman (1979; Table 1). These criteria are reproduced in **Table V-2** below.

Table V-2: Percent Tree Mortality Related to Defoliation Following Outbreak in Oregon and Washington Blue Mountains (reproduced from Wickman 1979).

Tree Defoliation Class (percent crown totally defoliated)	Percent Mortality Expected	
	Grand fir	Douglas-fir
1 (10%)	0	0
2 (25%)	1	4
3 (50%)	2	8
4 (75%)	7	5
5 (90%)	24	30
6 (99%)	53	46
7 (100%)	96	91

All treatment and control block data will be displayed by averaging current defoliation levels, and expected (2000 and 2001) tree mortality rates. Record all post-treatment defoliation and tree mortality estimation data on BIO. FORM NO. 10 (Refer to Appendix B).

A follow-up assessment of actual tree mortality in these same areas, including DFTM-caused, bark beetle-caused, and a combination of both, will be made in 2001 at the end of the bark beetle flight season to measure second year post-treatment tree mortality. All second-year (2001) follow-up tree mortality data will be recorded on BIO. FORM NO. 11 (refer to Appendix B). After the evaluation plots have been revisited and tree mortality measured and recorded for all plot trees, all flagging and tags will be removed from the plot site.