Section 9. Ozone Bioindicator Plants (West)

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9.1 OVERVIEW

Air pollutants, such as ground-level ozone, are known to interact with forest ecosystems. Ozone is the only regional gaseous air pollutant that is frequently measured at known phytotoxic levels (Cleveland and Graedel 1979; Lefohn and Pinkerton 1988). Ozone pollution has been shown to have an adverse effect on tree growth and alter tree succession, species composition, and pest interactions (Forest Health and Ozone 1987; Miller and Millecan 1971; Smith 1974). In addition, we know that ozone causes direct foliar injury to many species (Skelly and others 1987; Treshow and Stewart 1973). We can use this visible injury response to detect and monitor ozone stress in the forest environment. This approach is known as biomonitoring and the plant species used are known as bioindicators (Manning and Feder 1980). Ozone bioindicator plants are used to monitor changes in air quality across a region, and to assess the relationship between ozone air quality and Phase 2 and Phase 3 indicators of forest condition (e.g., growth increment and dieback).

A useful bioindicator plant may be a tree, a woody shrub, or a nonwoody herb species. The essential characteristic is that the species respond to ambient levels of ozone pollution with distinct visible foliar symptoms that are easy to diagnose. Field studies and/or fumigation experiments have identified ozone sensitive species and characterized the ozone specific foliar response for both eastern (Davis and Umbach 1981; Duchelle and Skelly 1981; Krupa and Manning 1988) and western (Richards and others 1968; Mavity and others 1995; Brace 1996) bioindicators. Foliar injury symptoms include distinct patterns of coloration, often associated with accelerated senescence.

This section describes procedures to select field sites for ozone biomonitoring using the FIA ozone grid, and to evaluate ozone injury on the foliage of sensitive plant species. Additional ozone sites, on an intensified ozone grid, may also be established by State and federal cooperators to improve the interpretive value of this indicator. This intensified sampling is done using the same methodology as the regular grid activities and is just as important.

9.1.1 SCOPE AND APPLICATION

The scope of this indicator is national, but procedures are amended regionally as needed, particularly with regard to suitable sites and target species. Other variables, such as number of species, number of plants, and methods of scoring are standardized nationally. The procedures, reporting, and assessment goals were developed with the following considerations:

- 1. Ozone plot distribution across the landscape covers both the more remote and expansive forests away from population centers and the more fragmented forests located in close proximity to urban areas;
- Ozone plot stratification nationwide reflects regional differences in air quality regimes and perceived risks to different forest types;
- 3. Sampling intensity in different regions is designed to allow links between ozone biomonitoring data and other FIA indicators;
- 4. Seasonal variability in ozone injury is addressed. We know that ozone injury must reach an undefined threshold within a leaf before the injury becomes visible to the human eye, and then tends to be cumulative over the growing season until fall senescence masks the symptoms.

NOTE: There are certain regions of the country where ambient ozone concentrations, during the growing season, routinely exceed levels that are known to injure sensitive plants. Other regions have relatively clean air. In regions with poor air quality, the crew data underscore the extent and severity of ozone pollution in the nation's forests. In regions with better air quality, the emphasis must be on establishing a baseline for the ozone indicator. In this regard, field crews that do not find ozone injury (zero values for the ozone injury variables) are making a significant contribution to the national FIA database.

9.1.2 SUMMARY OF METHOD

Field procedures include the selection of a suitable site for symptom evaluation, identification of three or more known ozone-sensitive species at the site, and identification of ozone injury on the foliage of up to 30 plants of each species. Each plant is evaluated for the percentage of injured area and severity of injury on a five-point scale. Field crews record information on the location and size of the opening used for biomonitoring, and record injury amount and severity ratings for each plant.

In the East, to eliminate problems with seasonal variability in ozone response, all foliar evaluations are conducted during a four-week window towards the end of the growing season. In the West, due to differences in growing season, topography, target species, and other regional factors that influence plant response to ozone, the identification of an optimum evaluation window for this indicator is problematic. Nevertheless, to maintain national consistency and improve crew logistics, the western regions use a midseason, five or six-week window for foliar injury evaluations.

In some States with a particular interest in air quality, foliar injury data are also collected from ozone sites on an intensified ozone grid. These supplementary ozone sites are standardized for certain site characteristics that influence ozone uptake by sensitive plants (Heck 1968; Krupa and Manning 1988), and are often co-located with physical air quality monitors. They are intended to improve the regional responsiveness of the ozone indicator.

Voucher specimens (pressed leaves with symptoms) are collected for each species for proper symptom identification. For each voucher, INJURY TYPE and INJURY LOCATION codes are recorded to fully describe the injury observed in the field. Additional quality control measures include field audits and remeasurement of 10% of the biomonitoring sites.

The implementation of an ozone grid independent of the traditional FIA plot system allows greater flexibility in plot location on the ground and greater sampling intensity in areas believed to be at high risk for ozone impact. In addition, plots are deliberately chosen for ease of access and for optimal size, species, and plant counts, thus maximizing data quality. Ozone is a regional pollutant, understood to have regional effects on vegetation. Therefore, data collected on the ozone grid will have direct application to the FIA P2 and P3 plots within the same region

No specialized safety precautions are necessary to complete the fieldwork for the ozone indicator.

9.1.3 SUMMARY OF PDR SCREENS AND TALLY PROCEDURES

Ozone indicator data are recorded on portable data recorders (PDRs). There are three data entry screens for ozone data: the Bioindicator Plot Identification Screen, the Plot Notes Screen, and the Bio Species Screen. On the handheld units, the corresponding screens are Plot Data, Ozone Notes, and Species Data. The Bioindicator Plot Identification Screen (Plot Data) includes a record of plot location and status as well as detail on site characteristics that influence ozone injury expression. The Plot Notes Screen (Ozone Notes) prompts crews to record safety tips and additional information that will help analysts interpret the results or assist subsequent crews collecting data at the same location. The Bio Species Screen (Species Data) prompts crews for injury AMOUNT and SEVERITY codes on a plant by plant basis. This screen includes a pop-up menu, which keeps a running total of numbers of plants and species evaluated by the field crews. Help screens may be accessed for any variable from any of the three data entry screens.

For a written summary of the data entry procedures, definitions, and codes for the ozone measurement variables refer to section 9.4 through 9.6.

9.1.4 EQUIPMENT AND SUPPLIES

- A large diameter, 10X hand lens for close examination of plant leaves for ozone injury.
- Reference photographs and laminated leaf samples to aid in symptom identification.
- A forester-grade plant press with cardboard inserts to store leaf vouchers collected in the field.
- Envelopes ready for mailing the leaf vouchers to the Western Regional Trainer
- Stiff paper or cardboard for protecting the leaf vouchers in the mailing envelopes.
- Flagging: for temporary marking of sites or sample plants.
- Three field data sheets: (1) For documenting Foliar Injury Data in the event of a PDR failure; (2) For preparing the plot location map; and (3) For recording Voucher Leaf Samples Data for QA. (see Appendix 9.B).

9.1.5 TRAINING AND QUALITY ASSURANCE

Each field crew member is trained and tested for familiarity with the site selection, species selection, and data collection procedures, and their ability to recognize ozone injury and discriminate against mimicking symptoms. Field crews are certified just prior to the beginning of the evaluation window for this indicator.

The National Ozone Advisor and one or more individuals in each region assume quality control responsibilities for the field season. Regional Advisors meet during a preseason session to refine methods and establish a unified approach to training, audits, and debriefing. Their responsibilities include (1) training and certifying the State trainers and/or field crews as needed for their region, (2) documenting hot audits of the field crews, (3) overseeing the field crew refresher session held just prior to the evaluation window for this indicator, (4) assisting in the field with remeasurement procedures for symptom quantification, and (5) conducting a debriefing for the indicator.

A field audit crew remeasures a subsample of the ozone ground plots in each region. Auditing procedures cover species selection, symptom identification, and quantification of injury, as well as foliar sample collection, preservation and shipment.

Results of the field audits and remeasurement activities are used to determine if the measurement quality objectives are being met. Regional Advisors and Field Supervisors who are certified for the ozone indicator have the authority to implement whatever corrective action is needed in the field (e.g., retraining and retesting).

9.1.6 VOUCHER SPECIMENS

Leaf samples are collected by field crews, cooperators, and all QA staff. They are to be placed in a small plant press immediately after removal from the selected plant. This is to preserve the integrity of the leaf sample and the injury symptoms until they can be validated by the National Indicator Advisor. A data sheet identifying the field crew and plot location is to be filled out and mailed with each sample.

Field crews, cooperators, and all QA staff collect leaf samples on the ozone biomonitoring sites according to procedures outlined in Subsection 9.2.7. These voucher specimens are pressed and mailed to the National Indicator Advisor for validation of the ozone symptom. If QA staff and regular field crews happen to be evaluating the same site at the same time, they collect and mail separate vouchers.

9.1.7 COMMUNICATIONS

Any questions arising during the field season that cannot be answered by the Field Supervisor or State Coordinator, should be directed to the National Indicator Advisor for the ozone indicator or to the Western Regional Trainer. If field crews try and are unable to reach the National Advisor or the Western Regional Trainer they may call the Regional Advisor for the North Central States, as indicated below. Keep in mind that Advisors may be in the field and, therefore, unavailable for phone calls during normal workday hours.

Messages left on answering machines should clearly identify who you are and when, where, and how to return your call. Please, be aware of differences in time zones and use email, if possible.

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9.2 OZONE BIOMONITORING PROCEDURES

NOTE: In the following discussion the words site, biosite, and plot are used interchangeably to refer to the open area used for the ozone biomonitoring evaluations.

The primary objective of the field crew procedures for the ozone indicator is to establish an ozone biomonitoring site within each polygon on the FIA ozone grid using the site selection guidelines provided in the Decision Table – section 9.2.2. These sites are used to detect and monitor trends in ozone air pollution

injury on sensitive species. Procedures include the selection of a suitable site for symptom evaluation, identification of three or more known ozone-sensitive species at the site, symptom identification and scoring on the foliage of up to 30 plants of each species, and the collection of voucher leaf samples. Each individual plant with ozone injury is scored for amount and severity of injury. Plants used for the selection of leaf vouchers are also evaluated for injury location and type. If a plant does not have ozone injury, it is still tallied with zeros for the amount and severity measurements. A hardcopy map, providing directions, plot coordinates, and key characteristics of the bioindicator site, is prepared for each plot.

All foliar evaluations are conducted during a mid-season ozone evaluation window. This helps address differences between plots that are caused by timing. During the window, all ozone sites on the ozone grid are evaluated for ozone injury. The same sites are evaluated every year.

9.2.1 EVALUATION WINDOW

Quantifying ozone injury on the FIA ozone plots is limited to an evaluation window starting in July and ending in mid-August. The evaluation window for crews in the Interior States begins the second week in July and extends through the third week in August. In the West Coast States, the window is open from the third week in July through the third week in August.

All established biomonitoring sites are evaluated each year. The ozone injury evaluations are generally completed over several weeks during the evaluation window depending on the size of the State and the number of crews dedicated to the ozone survey. Crews should adjust the timing of their evaluations for differences in elevation and latitude so that low elevation sites and/or more southern States use the earlier dates of the window while higher elevation sites and/or more northern States delay until the mid to later dates. Similarly, within each State, the low elevation, more southern biomonitoring sites should be evaluated first, the higher elevation, more northern sites last.

9.2.2 SITE SELECTION PROCEDURES

Candidate sites must be easily accessible open areas greater than one acre in size that are more than 100 feet (30 m) from a busy (paved) road. A site must contain at least 30 individuals of at least two bioindicator species to be evaluated for ozone injury. It is preferable that all sites have three or more species. The following table may be used as a decision guide for site selection:

Decision Table	First Choice = Best Site	Second Choice
Access:	Easy	Easy
Location:	Single location is used.	One or two locations (split-plot).
Size of opening:	>3 acres (1.2ha); wide open area; <50% crown closure.	Between 1-3 acres; long narrow or irregularly sized opening.
Species count:	More than three species.	Two or more species.
Plant count:	30 plants of 3 species;	30 plants of 2 species;
	10-30 plants of additional species.	10-30 plants of additional species.
Soil conditions:	Low drought potential. Good fertility.	Moderate dry. Moderate fertility.
Site disturbance:	No recent (1-3 years) disturbance;	Little or no disturbance;
	No obvious soil compaction.	No obvious soil compaction.

NOTE: In many parts of the West, the forested landscape is characterized by large natural openings populated by a single overstory species. Large areas with a single bioindicator species (e.g., aspen or ponderosa pine) may be selected for biomonitoring, but every attempt should be made to combine this single species site with a nearby location that includes one or more of the understory bioindicator species. Ozone is a regional pollutant, affecting large geographic areas, and sites within 3 miles of each other generally have the same ozone exposure regime.

The best ozone sites are often associated with wildlife preserves on public land. Private landowners are often eager to participate in the ozone program. State and county parks and wildlife openings also provide

good ozone sites. Other examples of suitable openings include old logging sites and abandoned pasture or farmland where you are reasonably certain that soil/site conditions are stable and free of chemical contaminants. Generally, if bedrock is exposed throughout an open area, then the soil conditions may be shallow, infertile, and often too dry to allow plants to respond to ozone stress. Sites that are routinely waterlogged are similarly unsuitable for biomonitoring. Avoid open areas where plants are obviously stressed by some other factor that could mimic or inhibit the ozone response. For example, the wooded edges of large parking lots in recreational areas are often highly compacted by car and foot traffic and should not be used. Do not select a site under a high-tension power line or on or near an active or reclaimed landfill. Do not select plants within 50 feet of the open edge around a cultivated field or tree plantation.

FIA crews and State Cooperators that have an established network of ozone sites may need to select and map replacement sites when previously mapped areas become overgrown or disturbed. Some sites may be split between two nearby locations to improve species and plant counts. In the case of split-plots, separate plot files (i.e., Tally files) are maintained for each location. Both have the same plot identification number (i.e., OZONE HEXAGON NUMBER) but different values for the plot number variable (i.e., OZONE PLOT NUMBER) as defined in Subsection 9.4.4.

No more than one half day should be spent locating a new bioindicator evaluation site. Crews must provide geographic coordinates (i.e., latitude and longitude) for all newly established ozone sites. If a site is split between two locations, the geographic coordinates for both locations are recorded.

NOTE: A split-plot consists of two different locations within 3 miles of each other, preferably with similar site characteristics. Species and plant counts from one location are combined with the species and plant counts from the second location to meet the species and plant count standards for site selection. On the PDR or data sheet, the same OZONE HEXAGON NUMBER is assigned to each location. However, each location is assigned a unique OZONE PLOT NUMBER; OZONE PLOT NUMBER = 1 for the first location that is evaluated by the field crew and OZONE PLOT NUMBER = 2 for the second location. In this way, separate Tally files are maintained for each location.

NOTE: The following table provides additional guidance on the required number of species and plants at each biomonitoring site. The best site has a minimum of 3 bioindicator species and 30 plants of each species. The best site may consist of two locations (split-plot) within 3 miles of each other if species or plants counts at any one location are low. If more than 3 species are found on a site, evaluate at least 10 plants of each additional species. For acceptable sites, establish a second location the following field season to increase species and plant counts. Marginal sites should be replaced the following field season.

Site Ranking	Number of species	Preferred number of plants per species	Acceptable number of plants per species	Minimum number of plants per species
Best	Species1	30	25 – 30	10
	Species2	30	10 – 30	10
	Species3	30	10 – 30	10
	Species4+	10 – 30	10 – 30	10
Acceptable	Species1	30	25 – 30	20
•	Species2	30	10 – 30	10
Marginal	Species1	30	25 – 30	25

9.2.3 SITE MAPPING

Once a bioindicator site is selected, the field crew records the estimated size of the site opening and other key site characteristics identified on the PDR or data sheet. The crew then maps the location of the site relative to some obvious and permanent marker such as a telephone pole, building, or property marker. Directions to the site, including road names and distances, are added to the map. Crews also mark the starting point for plant selection (see section 9.2.4) and approximate location of plant groupings used for

evaluation (see section 9.2.5) on the site map. If available, a GPS unit is used to determine plot coordinates and elevation. Otherwise, this information is obtained from a USGS topographic map, generally the $7\frac{1}{2}$ minute series quadrangle.

Ozone site maps are used by audit and regular crews in subsequent visits to the plot (see Figure 9-1) to ensure that the same site and the same population of plants are remeasured every year. This bioindicator site map must be kept with the appropriate state or federal cooperator so that it is readily available to whoever needs it.

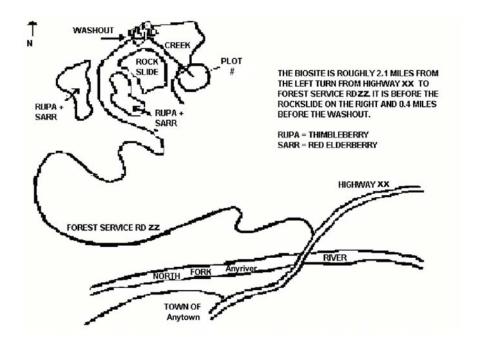


Figure 9-1. Example of a well-drawn map showing the location of the biosite and the approximate location of the bioindicator species and other key landmarks. Road names and North arrow are also included.

9.2.4 SPECIES SELECTION

At the selected bioindicator site, the crew evaluates 30 individuals of three or more bioindicator species. If three species cannot be found at the site, then a lesser number of species is still evaluated. Crews may combine species and plant counts from neighboring locations to obtain the required plant counts for each site. If 30 plants of two or more species cannot be found at the site, then a new site or additional location must be selected. A list of species is provided to the field crews for each region. Crews are encouraged to select from the top of the list down when several species are found at the same site. However, species with 30 or more individual plants should be a first priority for choice of species. Key identifying characteristics of each species are provided in the Appendix 9.A. Species ID information can also be accessed from the ozone indicator web site: fiaozone.net

Field crews record the species code number for each selected species in the PDR or on the data sheet. The target species and codes for each region are:

Interior Region

Code	Definition	Scientific Names
0122	Ponderosa pine ¹	Pinus ponderosa
0116	Jeffrey pine ²	Pinus jeffreyi
0960	Blue elderberry	Sambucus cerulea
0746	Quaking aspen	Populus tremuloides
0924	Scouler's willow	Salix scouleriana
0351	Red alder ²	Alnus rubra
0909	Skunk bush	Rhus trilobata
0905	Ninebark	Physocarpus malvaceus
0969	Mountain snowberry	Symphoricarpos oreaphilus
0907	Western wormwood	Artemesia ludoviciana
0961	Red elderberry	Sambucus racemosa
0965	Huckleberry	Vaccinium membranaceum
0968	Evening primrose	Oenothera elata

¹ Pinus ponderosa var. scopulorum (WY, CO)

West Coast Region

Wood Codet Region				
Code	Definition	Scientific Names		
0122	Ponderosa pine ¹	Pinus ponderosa		
0116	Jeffrey pine	Pinus jeffreyi		
0960	Blue elderberry	Sambucus cerulea		
0908	Mugwort	Artemesia douglasiana		
0746	Quaking aspen	Populus tremuloides		
0924	Scouler's willow	Salix scouleriana		
0351	Red alder	Alnus rubra		
0906	Pacific Ninebark (WC) ³	Physocarpus capitatus		
0909	Skunk bush	Rhus trilobata		
0905	Ninebark	Physocarpus mlvaceus		
0969	Mountain snowberry	Symphoricarpos oreap		
0907	Western wormwood	Artemesia ludoviciana		
0961	Red elderberry	Sambucus racemose		
0965	Huckleberry	Vaccinium membranaceum		
0968	Evening primrose	Oenothera elata		

¹ Pinus ponderosa var. ponderosa

9.2.5 PLANT SELECTION

After site and species selection, the next task is to contiguously sample 30 individual plants of each species. Thirty plants of a target species must be sampled if they are available on site. In fact, crews are strongly encouraged to evaluate 150 plants at each site (30 plants of five species), if possible. The value of the bioindicator data increases significantly with increased numbers of plants evaluated. This is true even if the crew records 30 consecutive zeros on three different species.

NOTE: The borders of some biomonitoring sites are difficult to determine and crews may be uncertain how much ground area to cover to complete the plant selection procedures. Specific guidelines are not set because the constraints on crew time and resources vary considerably from one State to the next.

² Pinus jeffreyi (NV); Alnus rubra (ID)

² WC = This species is only found west of the Cascades.

Time and safety concerns should take priority. Each crew must make every effort to maximize the number of plants and species evaluated for ozone injury at each plot location. Generally, ozone injury evaluations take 1 hour per site to complete and, assuming routine travel, crews are expected to complete two ozone sites in a ten-hour workday.

The following procedures help the crews to collect the bioindicator data in as systematic (i.e., unbiased) a way as possible.

- 1. Identify a starting point for plant selection. This point is mapped on the site data sheet so that audit and regular crews evaluate roughly the same population of plants in subsequent visits to the plot.
- 2. Move away from the starting point, towards the center of the opening.
- 3. Begin locating individuals in a sweeping pattern, selecting plants that are growing under the same or similar growing (microhabitat) conditions. Do not skip plants with little or no injury.
- 4. Select the more exposed plants (high sunlight exposure) and avoid suppressed and shaded individuals. Plants along the edge of an opening may be used if, in your judgment, they receive direct sunlight for three to four hours each day.
- 5. Avoid plants under 12 inches in height or so tall that at least half of the crown area cannot be seen or touched.
- 6. Evaluate the foliage that can be seen and touched on 30 plants of each species in the opening.
- 7. Record the amount and severity of injury for each plant evaluated (with or without symptoms) on the PDR or data sheet.

NOTE: A pop-up menu keeps track of the plant counts by species. For any one species, stop when the pop-up display indicates that 30 plants have been tabulated, or when no additional plants of that species can be found on site. 30 plants of 5 species can be tabulated or any combination of species and plants that adds up to 150 data line entries.

Some plants spread vegetatively. This means that neighboring plants are often genetically identical. To avoid repeat sampling of clonal material, take several steps between each plant selected for evaluation. Use a systematic approach to select individual plants. For example, select the plant closest to your left side then take several steps and select the plant closest to your right side and repeat. (A comparable systematic approach should be applied to all evaluated species to minimize bias in the plant selection process.) If it is difficult to distinguish individual plants or stems, use an approximate 2-foot square area to represent a single plant.

9.2.6 SYMPTOM IDENTIFICATION AND SCORING

The bioindicator species selected for each region are those that have been determined through field and laboratory studies to be highly sensitive to ozone air pollution. However, within a species, differences in genetics between individuals result in differential sensitivities to ozone. This means that often an individual of a species with severe air pollution injury is found growing immediately adjacent to another individual of the same species with few or no symptoms.

In addition to genetics, the age of the leaves (position on the stem, branch, or rosette) affects a plant's susceptibility to ozone air pollution. In general, leaves at 75% full expansion are the most sensitive and tend to show symptoms most definitively toward the center of the leaf. Older leaves show symptoms more widespread over the leaf surface, while younger leaves show symptoms more commonly near the leaf tip. If leaves on one branch are affected, then leaves at a similar leaf position on another branch should be

affected, especially for branches on the same side of the plant under similar environmental conditions (sun or shade leaves).

All of the western bioindicator species, except ponderosa and Jeffrey pine, have broad leaves. When scoring foliar symptoms on these broad-leafed plants, check for the following characteristics of ozone injury:

- Symptoms are more severe on mid-aged and older leaves. New leaves will have no or very little injury.
- Symptoms are most likely confined to the upper leaf surface, and are typically visible as tiny purple-red to black spots (stippling).
- Check leaves covering each other. Overlapped leaves will have no injury on the bottom leaf.
- There will be some uniformity to size and shape of the lesions (stippling) on a leaf.
- Later in the growing season, stippling may be associated with leaf yellowing or premature senescence. Check the ground for fallen leaves.

On ponderosa and Jeffrey pine, the most common needle symptom is chlorotic mottle. When scoring foliar symptoms on pines, check for the following characteristics of ozone injury:

- Symptoms are visible as <u>diffuse yellow areas</u> (chlorotic mottle) without sharp borders between green and yellow zones, on older needles. Not all needles in a fascicle will be uniformly affected.
- Chlorotic mottle is rarely seen on current-year needles except in high-ozone areas. On young needles it may appear more olive than yellow.
- Older needles that are directly exposed to sunlight may show the most severe chlorotic mottle. However, almost all exposed branches on a plant will be affected to some degree.
- Premature needle drop frequently occurs on ozone-injured pines, even on trees that do not show other ozone injury symptoms. Check for missing older annual whorls and for large numbers of needles on the ground. Live crowns may appear small and thin.

NOTE: Missing whorls on ponderosa pine should not be recorded as ozone injury without reliable evidence of other foliar injury symptoms, such as chlorotic mottle.

Each plant (broadleaf and conifer) with ozone injury is evaluated for the percent of the plant that is injured and the average severity of injury. For each plant located, the percentage of injured area and the severity of injury are both rated on a scale of 0 to 5 (see below). Both AMOUNT and SEVERITY estimates are confined to the exposed portion of the plant. If a plant does not have injury, it is still tallied with zeros for these measurements. For broad-leaved species, the AMOUNT and SEVERITY estimates are based on injury to the upper surface area of the leaves. For the pine species, examine all needle surfaces including the under sides, particularly if the needles have large amounts of winter fleck (NOT an ozone injury symptom) on the upper surfaces.

Percent Scale for Injury AMOUNT: Estimate and record the percentage of leaves (or needles) on the plant with ozone injury symptoms relative to the total number of leaves (or needles) on the plant.

CODE	DEFINITION
0	No injury; the plant does not have any leaves/needles with ozone symptoms.
1	1 to 6 percent of the leaves/needles have ozone symptoms.
2	7 to 25 percent of the leaves/needles are injured.
3	26 to 50 percent of the leaves/needles are injured.
4	51 to 75 percent of the leaves/needles are injured.
5	>75 percent of the leaves/needles have ozone symptoms.

Percent Scale for SEVERITY of Injury: Estimate and record the mean severity of symptoms on injured foliage.

CODE	DEFINITION
0	No injury; the plant does not have any leaves/needles with ozone symptoms.
1	On average, 1 to 6 percent of the leaf area of injured leaves/needles have ozone symptoms.
2	On average, 7 to 25 percent of the leaf area of injured leaves/needles have ozone symptoms.
3	On average, 26 to 50 percent of the leaf area of injured leaves/needles have ozone symptoms.
4	On average, 51 to 75 percent of the leaf area of injured leaves/needles have ozone symptoms.
5	On average, >75 percent of the leaf area of injured leaves/needles have ozone symptoms.

NOTE: Red and blue elderberry have compound leaves. Use the whole leaf, not each leaflet, to estimate injury AMOUNT and injury SEVERITY.

NOTE: The percent scale for ozone injury evaluations has a long history of application in plant disease research. The scale utilizes break points that correspond to the ability of the human eye to distinguish gradations of healthy and unhealthy leaf tissue (see Horsfall and Cowling 1978).

Proceed as follows:

- Record injury AMOUNT and injury SEVERITY ratings for each plant on the PDR or data sheet.
- 2. Use the notes section on the PDR or data sheet to add other information that will help interpret the results (e.g., below average rainfall for the area).
- 3. Collect a voucher leaf sample (three leaves of each injured species evaluated at each location) and mail them to the Western Regional Trainer using the guidelines presented in section 9.2.7.

NOTE: Foliar symptoms are easiest to see under overcast skies. Bright sun will make it difficult to see the ozone stipple or chlorotic mottle. Stand so that you reduce the glare on the leaf/needle surface. Long periods without rain will inhibit symptom development even on the most sensitive plants. If there is below average rainfall for the area, please note this in the PDR or on the data sheet.

9.2.7 COLLECTION OF LEAF SAMPLES AND VOUCHER DATA

The voucher leaf samples (leaves and/or needles) are a critical aspect of the data collection procedures as they provide the necessary validation of the ozone injury symptom observed in the field by the field crews. A plant press is essential to the collection of useable leaf samples and must be taken into the field by the field crews. Crew data that do not include a voucher leaf sample with a completed voucher data sheet are removed from the FIA database.

A voucher leaf sample must be collected for each injured species evaluated on the bioindicator site. For each injured, broad-leaved species, the voucher consists of three leaves that clearly show the ozone injury symptom. For pine species with ozone injury, the voucher consists of two small branches (small terminal or lateral branch containing the full complement of needles) with obvious chlorotic mottle. If a field crew records ozone injury on red alder, Scouler's willow, and ninebark then a minimum of one voucher (3 leaves) from each of the three species (9 leaves in all) is collected and mailed to the Western Regional Trainer. In this example, three voucher data sheets (one for each species) must be filled out and mailed with the leaf samples.

The most useful voucher leaf samples show obvious foliar injury symptoms. If injury symptoms are not obvious and severe, send whatever leaf sample is available even if it is only one leaf with faint symptoms. Cut the leaf at the petiole, shake off any excess moisture, and place the leaf on blotter paper in the plant press. Each leaf is placed in the press so that it does not overlap another leaf. Include a label with each leaf sample placed into the plant press that identifies which plot the sample came from (i.e., OZONE

HEXAGON NUMBER) and the date. Petiole labels are provided for this purpose. Record the information on the labels with indelible ink and then wrap them around the petiole of at least one leaf per sample

NOTE: Blue and red elderberry have compound leaves. Select the whole leaf (not individual leaflets) when preparing a voucher sample.

NOTE: If QA staff and regular field crews happen to be evaluating the same site at the same time, they collect and mail separate vouchers.

NOTE: The recognition of ozone injury symptoms in the field is not an exact science, and many other foliar injury symptoms can be mistaken for ozone injury. Crews are encouraged to collect voucher specimens of both known and suspected ozone injury in the field to send to the Western Regional Trainer for verification.

The voucher data sheet must be completed for plot identification codes (e.g., STATE, COUNTY, OZONE HEXAGON NUMBER and OZONE PLOT NUMBER), CURRENT DATE, CREW ID, CREW TYPE, SPECIES code(s). This sheet is filled out at the bioindicator site on the same day the sample is collected. In addition, the population of plants from which the leaf vouchers are selected must be evaluated by the field crews for INJURY LOCATION and INJURY TYPE (defined below), and for the amount of injury present on the leaf that is not ozone stipple. This information, together with the visible injury symptoms on the leaf samples, is used to validate the ozone injury data observed and recorded in the field by the field crews. For each species, the INJURY LOCATION and INJURY TYPE codes are intended to represent what the crew observed on the majority of the injured plants in the sample population. In contrast, the recorded estimates of percent injury caused by some stress other than ozone are based on what the crew observed on the injured leaf samples mailed in with the voucher data sheet.

The INJURY LOCATION and INJURY TYPE codes are recorded on the upper half of the voucher data sheet as follows:

INJURY LOCATION for Broad-leaved Species: Specify the leaf age or position of the leaves with ozone injury.

Code	Definition	
1	>50% of the injured leaves are younger leaves. Younger leaves are usually located towards the branch tip (e.g., aspen, willow, oak, ninebark, and huckleberry), or top of the plant (e.g.,	
2	elderberry, wormwood and snowberry). >50% of the injured leaves are mid-aged or older leaves. Mid-aged and older leaves are located	
۷	halfway along the branch (e.g., aspen, willow, oak, ninebark, and huckleberry) or main stem of the plant (e.g., elderberry, wormwood, and snowberry), or more towards the base of the branch or stem.	
3	Injured leaves are not concentrated in any one location, leaf age or position. Injury may be spread more or less evenly over the plant or is, otherwise, difficult to describe.	

INJURY LOCATION for Pines: Specify the leaf age or whorl with ozone injury.

Code	Definition
1	>50% of the injured needles are on the current whorl.
2	>50% of the injured needles are on whorls 1 year old and older.
3	Injury is not concentrated on any one needle whorl but is spread more or less evenly along the branch or is, otherwise, difficult to describe.

INJURY TYPE for Broad-leaved Species:	Specify the visible injur	v symptom

Code	Definition
1	The injury on >50% of the injured leaves is best described as upper-leaf-surface stipple (i.e., tiny purple-red to black spots occurring between the veins). Stippling may be associated with leaf yellowing and leaf drop late in the growing season; When injury is severe, stipples may coalesce and appear as uniform discoloration of the leaf surface.
2	The injury on >50% of the injured leaves is something other than upper-leaf-surface stipple. For example, small white to tan flecks occurring between the veins, or injury that is clearly visible on both leaf surfaces, or a general discoloration of the leaf that resembles early fall coloration.
3	The visible injury is varied or, otherwise, difficult to describe.

INJURY TYPE for Pines: Specify the visible injury symptom.

Code	Definition
1	The injury on >50% of the injured needles is best described as chlorotic mottle i.e., small patches of yellow tissue with diffuse borders and surrounded by apparently healthy (green) tissue. Chlorotic mottle may be associated with premature needle drop.
2	The injury on >50% of the injured needles is something other than chlorotic mottle. For example, winter fleck on the upper surface of the needles, or tipburn (i.e., reddish brown discoloration of the needle tips).
3	The visible injury is varied or, otherwise, difficult to describe.

NOTE: Not all location and type codes are indicative of ozone injury. Certain combinations of location and type codes, considered with a questionable leaf voucher, may invalidate the injury data. Other combinations provide quality assurance for the injury assessment. Crews should describe any unusual or questionable symptoms on the upper half of the voucher data sheet.

9.2.8 VOUCHER MAILING PROCEDURE

Vouchers are mailed in bulk at the end of the field season, or earlier, depending on the work schedule. It is very important to mail only dry, pressed leaf samples. Before mailing, make sure the upper half of the voucher data sheet has been completed. This sheet is filled out on the same day the sample is collected, even if the sample is not mailed on that day. Please comment on the weather or general plot conditions that might help interpret the injury data. For example, "It's been 14 days now without rain," "Every plant showed the same response and it was very obvious," or "This was a highly disturbed site."

NOTE: Crews are encouraged to add information on the biosite location to the voucher data sheet such as the uncoded name of the county or closest town. This helps the Western Regional Trainer map the initial findings from the leaf vouchers and alert FIA staff to high ozone areas.

The lower half of the voucher data sheet is filled out by the Western Regional Trainer to whom you are sending the sample. Place the voucher data sheet and the leaf sample between two pieces of stiff paper or cardboard before placing into a mailing envelope addressed to the Western Regional Trainer. Do not tape the leaves or needles to the paper or cardboard. Taped samples often break apart when they are handled, making evaluation difficult. Include as many samples as fit easily into each mailing envelope. There must be a unique voucher data sheet for each sample or species, unless you are using the form for multi-species. Keep leaf samples and the corresponding leaf voucher data sheets together. Leaf samples that are separated from the corresponding leaf voucher data sheets may be mislaid, especially if leaf labels are missing or incomplete.

NOTE: The Western Regional Trainer will make every effort to provide immediate feedback on the leaf vouchers. To facilitate this, crews must fill in the contact information on the voucher data sheet.

9.2.9 CREW MEMBER RESPONSIBILITIES

- Although one or two crew partners may be trained for this indicator, one person typically takes the lead responsibility for site selection, plant selection, and ozone injury evaluations. All procedures can be successfully completed by one person. Two person crews are recommended for safety reasons.
- 2. All members of the field crew may assist each other in the site selection process. Once a site is selected, one crew member is responsible for mapping the site and the location of bioindicator species on the field data sheet.
- 3. Only the crew member trained and certified in ozone injury evaluations may collect the amount and severity data and the leaf voucher. Other crew members may assist by recording the injury scores on the PDR or data sheet and by getting the plant press supplies ready.
- 4. The crew member that evaluates the plants for injury is responsible for collecting and mailing the voucher sample with air pollution symptoms.

9.2.10 FIELD PROCEDURES FOR UNTRAINED FIELD CREWS

There are certain procedures for the ozone indicator that may be performed by individuals that have not attended the ozone training and been certified to collect ozone data. These procedures still require some explanation and oversight by the certified crew member. Untrained personnel may assist in the selection and mapping of the ozone biomonitoring site and in the location and identification of bioindicator species on the selected site. They may not rate plant injury. It may also be helpful for the untrained crew person to act as the data recorder for the certified crew member, thus speeding up the data collection process.

9.3 SITE INTENSIFICATION

In addition to the unique ozone plots that are identified by the base ozone grid, some Cooperators have established additional biomonitoring sites to represent the local plant populations and environmental conditions. This is not an auxiliary effort, but an integral part of the monitoring activities for this indicator. In some States, additional biomonitoring sites are limited in number and are deliberately located close to weather and air quality monitoring stations. In other States, the ozone grid is intensified to allow for an unbiased allocation of additional biomonitoring sites. It is recommended that additional sites, whether few or many in number, be located on public land to facilitate the annual measurement activities.

Biomonitoring sites added to the base grid typically possess attributes of an ideal site for evaluating ozone injury on sensitive species. They are larger than three acres, contain the maximum number of indicator species, and have soil/site conditions with low drought potential and adequate fertility. They are evaluated for ozone injury using the same methods and during the same time frame as described in section 9.2. Voucher leaf samples must be collected, according to procedures described in section 9.2.7 and mailed to the Western Regional Trainer.

9.4 PLOT LEVEL DATA

All plot-level measurement codes for the ozone indicator are defined below. The codes and definitions are the same whether the crew is entering data using Tally (Paravant¹ or Husky) or a personal data assistant (Handspring or Palm).

¹ The use of trade or firm names in this publication is for reader information only and does not imply endorsement by the U.S. Department of Agriculture of any product or service.

Ozone plots vary in size and do not have set boundaries. When describing plot-level characteristics, use the predominant characteristics where most of the plant species are located. If conditions vary markedly across the site, or by species, then describe this in the plot notes or on the site map. Specify the elevation, aspect, terrain position, soil depth, soil drainage, and disturbance for the highest priority species (section 9.4) found on the site. The soil depth, soil drainage, and disturbance variables are intended to describe general conditions on the plot and are not based on actual measurements. For a complete explanation of the procedures associated with these measurement codes, refer to section 9.2.

9.4.1 STATE

Record the unique FIPS (Federal Information Processing Standard) code identifying the State where the plot center is located.

When collected: All plots Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time Values: See Appendix 1

9.4.2 COUNTY

Record the unique FIPS (Federal Information Processing Standard) code identifying the county, parish, Borough (or unit in AK) where the plot center is located.

When collected: All plots Field width: 3 digits Tolerance: No errors

MQO: At least 99% of the time Values: See Appendix 1

9.4.3 OZONE HEXAGON NUMBER

Record the unique code assigned to each ozone hexagon. In some cases this will be a former FHM or P3 hexagon.

When collected: All plots Field width: 7 digits Tolerance: No errors

MQO: At least 99% of the time

Values:

9.4.4 OZONE PLOT NUMBER

Record the plot number that describes whether an ozone plot consists of one or two locations. If two locations are selected, they must be within 3 miles of each other. Two locations are selected as needed to obtain optimal species and plant counts for each ozone plot. The OZONE HEXAGON NUMBER is the same for both locations.

When collected: All plots Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time

Values: 1 to 2

- 1 The ozone plot consists of a single location or this is the first location of a plot split between two locations.
- 2 The ozone plot is split between two locations. This code identifies the second location added by the field crew to increase species and plant counts for a single hexagon number.

9.4.5 QA STATUS

Record the code to indicate the type of plot data collected.

When collected: All plots Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time Values: 1 to 2 and 4 to 7

- 1 Standard ozone plot
- 2 Cold check
- 4 Training/practice plot (off grid)
- 5 Botched plot file
- 6 Blind check
- 7 Hot check (production plot)

9.4.6 CREW TYPE

Record the code to specify what type of crew is measuring the plot.

When collected: All plots Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time

Values: 1 to 2

- 1 Standard field crew
- 2 QA crew (any QA crew member present collecting remeasurement data)

9.4.7 OZONE SAMPLE KIND

Record the code that describes the kind of plot being visited. OZONE SAMPLE KIND has a value of 1 when an ozone plot is established in a previously empty polygon. OZONE SAMPLE KIND has a value of 2 when remeasurement occurs at the same location, or when the replacement plot is within 3 miles of the previously established plot. OZONE SAMPLE KIND has a value of 3 when the replacement plot is more than 3 miles away from the previously established plot.

When collected: All plots Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time

Values: 1 to 3

- 1 Initial plot establishment on the base grid or on a newly intensified grid.
- 2 Remeasurement of a previously established plot.
- 3 Replacement of a previously established plot that was replaced because the original plot could not be relocated or because it no longer met ozone plot measurement criteria.

9.4.8 CURRENT DATE

Record the year, month, and day that the current plot visit was completed as follows:

9.4.8.1 YEAR

Record the year that the plot was completed.

When collected: All plots Field width: 4 digits Tolerance: No errors

MQO: At least 99% of the time

Values: Beginning with 1998, constant for a given year

9.4.8.2 MONTH

Record the month that the plot was completed.

When collected: All plots Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time

Values:

January	01	May	05	September	09
February	02	June	06	October	10
March	03	July	07	November	11
April	04	August	80	December	12

9.4.8.3 DAY

Record the day of the month that the plot was completed.

When collected: All plots Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time

Values: 01 to 31

9.4.9 OZONE GRID DENSITY

Record the code that identifies whether the plot is on the base ozone grid or on an intensified ozone grid. Note: The OZONE GRID DENSITY value = 2 when there are two ozone plots with different OZONE HEXAGON NUMBERS in the same polygon.

When collected: All plots Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time

Values: 1 to 2

Unique ozone plot within a polygon. (1 site:1polygon)One of two or more ozone plots within the same polygon

9.4.10 PLOT SIZE

Record the code that indicates the size of the opening used for biomonitoring.

When collected: All plots Field width: 1 digit Tolerance: No errors MQO: Repeatable estimate Values: 1 to 2

- 1 Greater than three acres.
- 2 Greater than one acre, but less than three acres.

9.4.11 ASPECT

Record the code that identifies the direction of slope for land surfaces with at least 5 percent slope as measured with a hand compass to the nearest degree.

When collected: All plots Field width: 3 digits Tolerance: +/- 30°

MQO: At least 99% of the time

Values:

000 No aspect, slope < 5 percent

001 1 degree002 2 degrees

. .

360 360 degrees, due north

9.4.12 TERRAIN POSITION

Record the code that identifies the position of the plot in relation to the surrounding topography.

When collected: All plots Field width: 1 digit

Tolerance: Repeatable estimate MQO: At least 99% of the time

Values: 1 to 5

- 1 Ridge top or upper slope
- 2 Bench or level area along a slope
- 3 Lower slope
- 4 Flat land unrelated to slope
- 5 Bottom land with occasional flooding

9.4.13 SOIL DEPTH

Record the code that indicates the general depth of the soil where most of the bioindicator species are growing.

When collected: All plots Field width: 1 digit

Tolerance: Repeatable estimate MQO: At least 99% of the time

Values: 1 to 2

- 1 Bedrock is not exposed.
- 2 Bedrock is exposed; Soil is generally shallow.

9.4.14 PLOT WETNESS

Record the code that identifies the degree of wetness where most of the bioindicator plants are growing.

When collected: All plots Field width: 1 digit

Tolerance: Repeatable estimate MQO: At least 99% of the time

Values: 1 to 3

1 This is a wet plot; Riparian zone or bottomland.

2 This plot is moderately dry; Meadow or Northeast facing slope.

3 This plot is very dry; Exposed ledge, desert or alpine area.

9.4.15 DISTURBANCE

Record the code that identifies the presence and kind of disturbance where most of the bioindicator plants are growing. The area affected by any human caused or natural disturbance must be clearly visible and recent enough to influence plant health and condition. Disturbance that results in significant soil compaction is especially significant.

When collected: All plots Field width: 1 digit

Tolerance: Repeatable estimate MQO: At least 99% of the time

Values: 0 to 2

0 No recent or significant disturbance.

1 Evidence of overuse; Human activity causing obvious soil compaction or erosion.

2 Evidence of natural disturbance including fire, wind, flooding, grazing, pests, etc.

9.4.16 INJURY CHECK

Record the code that indicates whether ozone injury was observed on non-tallied plants or species. This variable allows a plot to be identified as impacted by ozone even though there is no quantitative data on injury severity for trend analyses. A leaf voucher must be collected to validate the injury.

When collected: All plots Field width: 1 digit Tolerance: No error

MQO: At least 99% of the time

Values: 0 to 1

0 No injury was observed on non-tallied plants or species.

1 Ozone injury was observed on non-tallied plants or species and a leaf voucher collected.

9.4.17 ELEVATION

Obtain elevation data from USGS topographic maps, generally the $7\frac{1}{2}$ minute series quadrangle. Locate the area where most of the bioindicator species are growing and record elevation to the nearest foot.

When collected: When GPS UNIT = 0

Field width: 6 digits Tolerance: +/-200 feet

MQO: At least 99% of the time

Values:

9.4.18 Plot Notes

Use these fields to record notes pertaining to the entire plot. If the notes apply to a specific aspect of the plot, then make that clear in the notes. Record the location where GPS coordinates were collected, and GPS file name, as needed. If no GPS Unit was available, record the geographic coordinates (i.e., latitude and longitude) of the plot center in Degrees, Minutes, and Seconds using USGS topographic maps, generally the 7½ minute series quadrangle.

9.4.18.1REMARK1 and REMARK2

Record any information on site characteristics, use of supplemental species, safety, plant location, injury patterns, or recent rainfall amounts that will assist subsequent crews visiting the site or help interpret the results.

When collected: All plots

Field width: Unlimited alphanumeric character field

Tolerance: N/A MQO: N/A

Values: English language words, phrases and numbers

9.5 GPS COORDINATES

Use a global positioning system (GPS) unit to determine the plot coordinates and elevation of all ozone plot locations. GPS readings are collected according to procedures outlined in the FIA National Core Field Guide for Phase 2 & 3 Plots, Version 2.0. The ozone data entry applications accept GPS readings obtained using a geographic coordinate system (not UTM). If you are using UTM, record readings on the field data sheet for mapping and on the PDR Plot Notes screen. If GPS coordinates cannot be collected, elevation and plot coordinates are obtained from USGS topographic maps, generally the 7½ minute series quadrangle. Record ELEVATION on the Plot ID screen and approximate latitude and longitude on the Plot Notes screen.

Use a global positioning system (GPS) unit to determine the plot coordinates and elevation of all field-visited plot locations.

NOTE: For several of the following GPS variables, the term plot center is used. There may be no obvious center to the ozone plots. Coordinates are collected as close as possible to a central location or marker that clearly locates the plot for returning crews. Explanatory notes are added to the plot map and Plot Notes screen as needed.

9.5.1 GPS Unit Settings, Datum, and COORDINATE SYSTEM

Consult the GPS unit operating manual or other regional instructions to ensure that the GPS unit internal settings, including Datum and Coordinate system, are correctly configured.

Each FIA unit will determine the Datum to be used in that region. Most will use the NAD 27 Datum (also known as NAS-C or NA 27 CONUS/CLK66), but coordinates collected using any appropriate datum can be converted back to a national standard (NAD 83) for reporting purposes.

Each FIA unit will also determine which coordinate system to use. Regions using a Geographic system will collect coordinates in Degrees, Minutes, and Seconds of Latitude and Longitude; the regions using the UTM coordinate system will collect UTM Easting, Northing, and Zone.

9.5.2 Collecting Readings

Collect at least 180 GPS readings at the plot center (see Note above). These may be collected in a file for post-processing or may be averaged by the GPS unit. Each individual position should have an error of less than 70 feet if possible (the error of all the averaged readings is far less).

Soon after arriving at plot center, use the GPS unit to attempt to collect coordinates. If suitable positions (180 readings at error less than or equal to 70 feet) cannot be obtained, try again before leaving the plot center.

If it is still not possible to get suitable coordinates from plot center, attempt to obtain them from a location within 200 feet of plot center. Obtain the azimuth and horizontal distance from the "offset" location to plot center. If a PLGR unit is used, use the Rng-Calc function in the PLGR to compute the coordinates of the plot center. If another type of GPS unit is used, record the azimuth and horizontal distance as described in Sections 1.14.12 and 1.14.13.

Coordinates may be collected further away than 200 feet from the plot center if a laser measuring device is used to determine the horizontal distance from the "offset" location to plot center. Again, if a PLGR unit is used, use the Rng-Calc function in the PLGR to compute the coordinates of the plot center. If another type of GPS unit is used, record the azimuth and horizontal distance as described in Sections 1.14.12 and 1.14.13.

In all cases try to obtain at least 180 positions before recording the coordinates.

9.5.3 GPS UNIT

Record the kind of GPS unit used to collect coordinates. If suitable coordinates cannot be obtained, record 0.

When collected: All field visited plots

Field width: 1 digit Tolerance: No errors

MQO: At least 99% of the time

Values: 0 to 4

- 0 GPS coordinates not collected
- 1 Rockwell Precision Lightweight GPS Receiver (PLGR)
- 2 Other brand capable of field-averaging
- 3 Other brands capable of producing files that can be post-processed
- 4 Other brands not capable of field-averaging or post-processing

9.5.4 GPS SERIAL NUMBER

Record the last six digits of the serial number on the GPS unit used.

When collected: When GPS UNIT >0

Field width: 6 digits
Tolerance: No errors

MQO: At least 99% of the time Values: 000001 to 999999

9.5.5 GPS DATUM

Record the acronym indicating the map datum that the GPS coordinates are collected in (i.e., the map datum selected on the GPS unit to display the coordinates).

When collected: When GPS UNIT >0 Field width: 5 characters (cccnn)

Tolerance: No errors

MQO: At least 99% of the time

Values:

NAD27 North American Datum of 1927 NAD83 North American Datum of 1983 WGS84 World Geodetic System of 1984

9.5.6 Latitude

Record the latitude of the plot center to the nearest hundredth second, as determined by GPS.

NOTE: The following can be customized at the region level (e.g., decimal minutes to the nearest thousandth) as long as the final results recorded are within the specified tolerance to the nearest hundredth of a second or +/- 1.01 ft.

9.5.6.1 LATITUDE DEGREES

Record the latitude degrees of the plot center as determined by GPS.

When collected: When GPS UNIT = 1, 2, 3 or 4

Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time

Values:

9.5.6.2 LATITUDE MINUTES

Record the latitude minutes of the plot center as determined by GPS.

When collected: When GPS UNIT = 1,2,3 or 4

Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time

Values: 1 – 59

9.5.6.3 LATITUDE SECONDS

Record the latitude decimal seconds of the plot center to the nearest hundredth place as determined by GPS.

When collected: When GPS UNIT = 1, 2, 3 or 4

Field width: 4 digits Tolerance: +/- 140 ft

MQO: At least 99% of the time

Values: 0.00 - 59.99

9.5.7 Longitude

Record the longitude of the plot center to the nearest hundredth second, as determined by GPS.

NOTE: The following can be customized at the region level (e.g., decimal minutes to the nearest thousandth) as long as the final results recorded are within the specified tolerance to the nearest hundredth of a second or +/- 1.01 ft.

9.5.7.1 LONGITUDE DEGREES

Record the longitude degrees of the plot center as determined by GPS.

When collected: When GPS UNIT = 1, 2, 3 or 4

Field width: 3 digits Tolerance: No errors

MQO: At least 99% of the time

Values:

9.5.7.2 LONGITUDE MINUTES

Record the longitude minutes of the plot center as determined by GPS.

When collected: When GPS UNIT = 1, 2, 3 or 4

Field width: 2 digits Tolerance: No errors

MQO: At least 99% of the time

Values: 1 - 59

9.5.7.3 LONGITUDE SECONDS

Record the longitude decimal seconds of the plot center to the nearest hundredth place as determined by GPS.

When collected: When GPS UNIT = 1, 2, 3 or 4

Field width: 4 digits
Tolerance: +/- 140 ft

MQO: At least 99% of the time

Values: 0.00 - 59.99

9.5.8 GPS ELEVATION

Record the elevation above mean sea level of the plot center, in feet, as determined by GPS. If no GPS Unit is available, record elevation from the appropriate USGS topographic map.

When collected: When GPS UNIT = 1, 2 or 4

Field width: 6 digits

Tolerance:

MQO: At least 99% of the time Values: -00100 to 20000

9.5.9 GPS ERROR

Record the error as shown on the GPS unit to the nearest foot.

When collected: When GPS UNIT = 1 or 2

Field width: 3 digits Tolerance: No errors

MQO: At least 99% of the time Values: 000 to 070 if possible

071 to 999 if an error of less than 70 cannot be obtained

9.5.10 NUMBER OF GPS READINGS

Record a 3-digit code indicating how many readings were averaged by the GPS unit to calculate the plot coordinates. Collect at least 180 readings if possible.

When collected: When GPS UNIT = 1 or 2

Field width: 3 digits Tolerance: No errors

MQO: At least 99% of the time

Values: 001 to 999

9.5.11 GPS FILENAME (CORE OPTIONAL)

Record the filename containing the GPS positions collected on the plot.

When collected: When GPS UNIT = 3

Field width: 8 characters.3 characters e.g. R0171519.ssf

Tolerance: No errors

MQO: At least 99% of the time Values: Letters and numbers

9.6 FOLIAR INJURY DATA

All measurement codes for the foliar injury data are defined below. Plants selected for ozone injury evaluations are rated for the percent of injured area and the severity of injury on a scale of 0 to 5 (see section 9.2.6). If a plant does not have injury, it is tallied with zeros for these measurements. A pop-up menu keeps track of plant counts by species. The plot is complete only when 30 plants of at least 3 species have been tallied, or when no additional plants can be found on the plot. Ozone plots vary in size and do not have set boundaries. Time and safety concerns should dictate how much ground area to cover to complete the foliar injury evaluation procedures.

9.6.1 SPECIES

Record the three-digit code that identifies each species on the plot. Species codes may be entered in the order they are encountered as you walk through the plot evaluating plants. A popup menu keeps a running total of numbers of plants and species evaluated.

When collected: All plots Field width: 4 digits Tolerance: No error

MQO: At least 90% of the time

Values: See 9.2.4

9.6.2 AMOUNT

Record the code that identifies the percentage of leaves on the plant with ozone injury symptoms relative to the total number of leaves on the plant. The percent scale code and definitions are fully described in section 9.2.6.

When collected: All plots Field width: 1 digit Tolerance: +/- 1 class

MQO: At least 90% of the time

Values: 0 - 5

- 0 No injury; The evaluated plant does not have any leaves or needles with ozone symptoms.
- 1 1 to 6 percent of the leaves/needles have ozone symptoms
- 2 7 to 25 percent of the leaves/needles are injured.
- 3 26 to 50 percent of the leaves/needles are injured.
- 4 51 to 75 percent of the leaves/needles are injured.
- 5 Greater than 75 percent of the leaves/needles have ozone symptoms.

9.6.3 NUMBER OF PLANTS

Record the number of plants tallied so far with no injury. When 0 is entered for AMOUNT, the PDR prompts for the NUMBER OF PLANTS with no injury. When a number greater than zero is entered for AMOUNT, the PDR prompts for the associated SEVERITY value. Zero and non-zero values for any species can be entered as they are encountered on the plot. The pop-up menu keeps track of plant counts by species.

When collected: When AMOUNT = 0

Field width: 2 digits Tolerance: No error

MQO: At least 90% of the time

Values: 1 to 30

9.6.4 SEVERITY

Record the code that identifies the mean severity of symptoms on injured foliage. The percent scale code and definitions are fully described in section 9.2.6.

When collected: When AMOUNT > 0

Field width: 1 digit Tolerance: +/- 1 class

MQO: At least 90% of the time

Values: 0 - 5

- 0 No injury. The plant does not have any leaves or needles with ozone symptoms.
- 1 On average, 1 to 6% of the leaf area of injured leaves/needles has ozone symptoms.
- 2 On average, 7 to 25% of the leaf area of injured leaves/needles has ozone symptoms.
- 3 On average, 26 to 50% of the leaf area of injured leaves/needles has ozone symptoms.
- 4 On average, 51 to 75% of the leaf area of injured leaves/needles has ozone symptoms.
- 5 On average. >75% of the leaf area of injured leaves/needles has ozone symptoms.

9.7 REFERENCES

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9.8 ACKNOWLEDGEMENTS

The National Advisor for the ozone indicator wishes to thank the individuals within FIA and FHM, as well as those outside the Forest Service that took the time to review this training section and offer suggestions for improvement and essential information to complete the guide. Special thanks to Pat Temple for his contributions to the text and to Pat, Dan Duriscoe, John Pronos, David Karnosky, Robert Kohut, and Dave Peterson who provided slides demonstrating ozone injury symptoms on the target bioindicator species for the western FIA regions. The National Advisor for this indicator may be contacted at: Gretchen Smith, Department of Natural Resources Conservation, 160 Holdsworth Way, University of Massachusetts, Amherst, MA 01003-4210 or via email at gcsmith@forwild.umass.edu

Appendix 9.A Key Identifying Characteristics of the Ozone Bioindicator Species

- 1. Ponderosa Pine is a large tree, up to 230 feet in height. Young tree bark is often thin and dark brown to black. Older tree bark is thick becoming yellow-red to cinnamon red and forming plates which slough off freely. Needles in bundles of three, 5-10 inches in length, not glaucous and yellow-green in color. Buds are resinous with redbrown scales and dark-hairy. Cones with a prickle at the tip of each scale. May be confused with Jeffrey pine which differs by having non-resinous, light-brown buds, and grayish blue-green glaucous needles.
- 2. **Jeffrey Pine** is a smaller tree than ponderosa pine, with darker cinnamon-red bark that may be tinged with lavender on old trunks. Needles in bundles of three, 5-10 inches in length, blue-green, and somewhat twisted. Crushed needles and twigs have a violet-like or pineapple odor. Buds are never covered with resin droplets. Cones with a prickle at the tip of each scale. May be confused with ponderosa pine.
- 3. **Quaking Aspen** is a medium sized tree up to 118 feet in height. Bark is smooth, greenish-white. Buds shiny but not resinous. Leaf petiole is strongly flattened. The leaf blade is broadly ovate (almost round) with a tapering tip and finely toothed margins, upper surface smooth, lower surface covered with a bloom. Aspen could be confused with black cottonwood which differs in its resinous buds, rough bark and round leaf petioles.
- 4. **Scouler's Willow** is a small tree or shrub up to 32 feet in height. Leaf blade is 1-4 inches in length, narrowly elliptic with the widest portion toward the tip, entire to irregularly toothed margins, lower surface smooth, upper surface shiny. This willow is NOT restricted to riparian zones. It can be easily confused with a number of other willow species. The combination of leaves widest toward the tip (mostly rounded ends and narrowly tapered bases) and the tolerance for upland (drier) habitats makes this willow relatively easy to identify.
- 5. **Pacific Ninebark** is a deciduous shrub 6-12 feet in height. Leaves alternate, 3 or 5 lobed (maple-like), 2-3 inches long, serrate, dark green and smooth above, paler and hairy below. Twigs red to grayish brown, splits longitudinally into long strips. Flowers small, white, borne in a cluster, stems hairy. Very similar to ninebark (see below) which is generally smaller, in drier habitats, and with densely hairy ovaries.
- 6. **Ninebark** is an erect, loosely branched shrub with maple-like leaves and shreddy bark. May be up to 6 feet in height. Leaves and flowers similar to Pacific ninebark except the ovaries are densely hairy. May be confused with Douglas maple which has opposite leaves, or sticky currant, which has leaves that are sticky to the touch. Often associated with ponderosa pine and Douglas-fir forests at low to mid-elevation.
- 7. **Huckleberry** is an erect shrub 3-5 feet high. Leaves 1-2 inches long, half as wide, thin and pale green on both surfaces, smooth or occasionally minutely hairy, margins toothed, apex and base both acute. Fruit deep purple to black round berry around 6 mm diameter. Twigs slender, green and ridged. Found on dry to moist sites, sun or shade. Similar, and often found with oval-leaved huckleberry which has entire (smooth) rather than toothed leaves.
- 8. **Blue Elderberry** is a tall deciduous shrub, sometimes tree-like, up to 20 feet in height. Twigs with a soft pith inside. Leaves opposite, pinnately compound, the 5-9 leaflets sharply serrate and strongly uneven at the base. Flowers small, white, flat-topped cluster. Fruit a blue-black berry covered with a white powdery bloom. This species could be confused with red elderberry which differs by having flowers in a spike and red-purple fruit. Found mostly on moist, well-drained sites in the sun; sea level to 9,000 ft.
- 9. **Red Elderberry** is a tall deciduous shrub, sometimes tree-like, up to 20 feet in height. Twigs with a soft pith inside. Leaves opposite, pinnately compound, the 5-7 leaflets sharply toothed and often uneven at the base. Flowers small, white, and clustered into a long spike. Fruit is a berry, most often red in color but sometimes purplish-black or yellow. Similar to blue elderberry which has a flat-topped flower cluster and a blue-black berry.
- 10. Western Wormwood is an aromatic perennial herb, 1-3 feet in height. Leaves mostly 1-4 inches long, variable in shape but most often with 3-5 narrow lobes, white hairy beneath, sometimes above as well. Flowers small and arranged in a loose, narrow flower cluster, 2-12 in long. May be confused with Douglas' wormwood which has wider leaves and is usually found in moister habitats. Also similar to Riverbank wormwood which occurs only near streams and outwash areas.
- 11. *Mugwort* is a large perennial herb 2-5 feet tall, usually found in large colonies in wet areas, ditches, or drainages. Leaves are evenly-spaced, 0.4-4.0 inches long, the upper leaves are narrowly elliptical, the lower widely oblanceolate, often coarsely 3 to 5 lobed near the leaf tip, 0.8-1.0 inch wide, green above, covered with dense white hair beneath. Differs from western wormwood in having wider lower leaves and in its generally damp habitat.

- 12. **Evening Primrose** is a large biennial with elliptical leaves up to 10 inches long in a dense rosette the first year. The large (>1m) flowering stalk with long red-tinged elliptical leaves and large bright yellow four-petaled flowers forms in the second year. Both the leaves and stem are densely hairy, and the hairs often have red, blister-like bases. Usually found in moist, sunny habitats, like seeps or meadows.
- 13. **Mountain Snowberry** is a shrub, 1.5-5 feet in height with a solid brown pith. Bark: shreddy, brownish. Young twigs: hairy. Leaves opposite, elliptical, 0.4-1.4 inches long and half as wide. Flowers (May-June) tubular-shaped, the petals white with a pink tube. Fruit a white berry. Common snowberry differs by having non-tubular flowers and a hollow pith. Trailing snowberry is a trailing shrub with non-tubular flowers; and Utah honeysuckle has larger leaves and a solid white pith.
- 14. Red Alder is a deciduous tree up to 65 feet tall with dark green leaves 2.4-4.7 inches long. The leaves are coarsely toothed, with smaller teeth on the leaf margins, and the leaf veins are also tightly inrolled. Red alder is a common tree in damp situations and is a frequent colonizer of clearings, especially following clearcuts in coniferous forests.
- 15. **Skunkbush** is a small, diffusively-branched shrub, 1.6-3.3 feet tall. The tips of the branches often droop down almost to ground level. The leaves are alternate, compound, with three leaflets, each of which is 3-lobed. The leaves resemble those of poison oak, but the leaflets of skunkbush are smaller, more hairy, and much more deeply-lobed. The leaves of skunkbush also emit a strong, ill-scented odor when crushed. However, if unsure, DO NOT crush the leaves of a shrub with three leaflets to determine the odor. Skunkbush is usually found on dry, open, brushy hillsides, while poison oak prefers damp or shaded forested areas and riparian habitats. Skunkbush is found throughout the southwest, from California and Arizona north to Colorado and Idaho.

Appendix 9.B Data Sheets

OZONE BIOINDICATOR PLANTS

BioSite Characteristics – West

This sheet must be completed only if you have not entered this same information on the Bioindicator Plot ID screen.

To be	filled out	by the FIELD	CREW or Co	ooperator:	Refer to	Field Guid	le for co	ode defin	itions.

State	County	Ozone Hexagon Number	O3Plot	Number ¹	Month	Day	Year	Crew ID	Crew Type
									Regular QA
¹O3Plo	ot Number re	fers to the number of locations (1 o	r 2) used fo	r each hexago	n number. A	separat	e sheet sho	uld be used for	each location.
Ozon	Ozone Sample Kind: Initial biosite establishment on the FIA ozone grid. (Data collection in a previously empty polygon)								
				-					,
		rement of a previously establis					-		
	Replacen	nent of a previously established	1 biosite th	at was repla	iced to mee	t site sel	ection gu	idelines (or lo	ost site).
Ozon	e Grid Dei	nsity: (Is the grid intensified	d, or not?)					
	This hex	number identifies a unique	ozone bi	osite within	n a polygo	n (1 site	e:1 polyg	on)	
	One of t	wo or more ozone biosites w	vithin the	same poly	gon, each	with the	eir own h	exagon nun	nber.
	•							· ·	
√ Plea	ase put a c	heck mark beside the correc	ct informa	ation. Pleas	se complet	e all da	ta fields.		
BioSi	te size: (P	lot Size)	,	Terrain po	sition:				
	> 3.	0 acres (1.2 hectares)			Ridge to	p or up	per slop	e	
	1to	3 acres (0.4 – 1.2 hectares)						g a slope	
	Oth	er: please describe			Lower s	lope			
					Flat land	d unrela	ated to sl	оре	
					Bottom	land wi	th occasi	onal floodin	g
Aspec	ct: 000° = 1	no aspect; 360° = N aspect]	Elevation:	record est	imate ii	ı feet or	meters	
Reco	rd to neare	est degree =]	Feet = Meters =					
BioSi	te Wetness	(PlotWet):		Soil Drai	nage: [opti	on] S	Soil Dept		
		Ex: riparian zones, botton		Well-drained Bedrock not expose					
		tely dry Ex: meadow, NE s		W			В	edrock expo	sed
	Very dr	y Ex: exposed ledge, desert	t, alpine.	Ex	cessively (lry			
Distu		isturbance on the site or in					_	are growing	•
		nt or significant disturbance							
		e of overuse; Human activit							
	Evidenc	e of natural disturbance inc	luding fir	e, wind, flo	oding, gr	azing, p	ests, etc.		
Fill in	ı below all	that apply. Check here if ge	eographic	coordinate	es were ob	tained i	from a to	ppographic i	тар:
GPS.	Type:	Datum:		GPS	Serial Nui	nher:			
Latitu	• •	Dutumi			Error =	110011			
	itude =				ber of Rea	dings =			
	Elevation = GPS File Name =								
Easti		Northing:			ror(ft.):	·	G	rid Zone:	
		nde information on additional spec	ies in the ar	•	, ,	dditional			nay be useful.

 $File \ this \ completed \ data \ sheet \ with \ the \ bioindicator \ site \ map \ and \ driving \ directions \ in \ the \ ozone \ plot \ files \ for \ your \ State \ or \ Region.$

3.0 Phase 3 Field Guide - Ozone Bioindicator Plants (west) October, 2005

OZONE BIOINDICATOR FOLIAR INJURY DATA SHEET West Coast and Interior West and Plain States

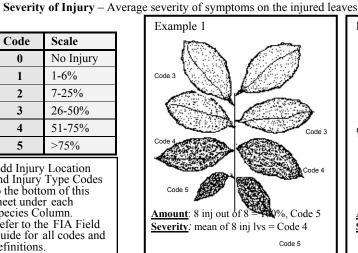
State	County	O3HexagonNum	O3PlotNum	Month	Day	Year	Measurement Type
							Reg. Crew/Cooperator QA crew

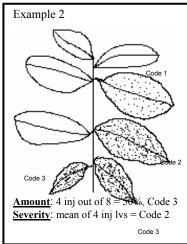
Code	Species
122	Ponderosa pine
746	Quaking aspen
924	Scouler's willow
116	Jeffrey pine
351	Red alder
969	Snowberry spp.
905	Ninebark
906	Pacific Ninebark
965	Huckleberry
960	Blue elderberry
961	Red elderberry
907	Western wormwood
908	Mugwort
909	Skunk bush
968	Evening primrose

Amount of Injury – % of leaves injured relative to the total leaf number

Code	Scale
0	No Injury
1	1-6%
2	7-25%
3	26-50%
4	51-75%
5	>75%

Add Injury Location and Injury Type Codes to the bottom of this sheet under each Species Column. Refer to the FIA Field Guide for all codes and definitions.





	Species Code		Species Code Species Code		Species Code			Species Code			Species Code	
Plant	Amount	Severity	Amount	Severity	Amount	Severity		Amount	Severity		Amount	Severity
1						j			ĺ			
2												
3												
4												
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25												
26												
27												
28												
29												
30	İ			1	İ				İ			l

Did you collect 3 leaves that clearly show ozone stipple, for each injured species?

Month Day

Crew ID

OZONE BIOINDICATOR PLANTS

Data Sheet for Directions and Mapping for the Bioindicator Site Location

To be filled out by the FIELD CREW or Cooperator: Refer to Field Guide for code definitions.

Ozone Hexagon Number O3Plot Number¹

¹ O3Plot N location	Number refers to	the number of	locations (1 or 2) u	ised for each	hexagon nun	nber. A sepa	rate sheet	should be	used for each	
location	•									

Include the following information on the map:

- 1. Location of the site relative to some obvious and permanent marker.
- 2. Road names and distances as needed.
- 3. North arrow.
- 4. Species codes and approximate location of plant groupings used for the ozone injury evaluations.
- 5. Location and distance to two major roads; distance and direction to two major towns.
- 6. Gazetteer reference page if available.

Return the original of this map to the corresponding Biosite Folder so that it can be used by audit and regular crews in subsequent visits to the plot. Mail a copy to the National Indicator Advisor the year that the site is established.

GPS Type:	Datum =	GPS Serial Number:	
Latitude =		GPS Error =	
Longitude =		Number of Readings =	
Elevation =		GPS File Name =	
Easting:	Northing:	+/-Error(ft.):	Grid Zone:

OZONE BIOINDICATOR PLANTS General Information

Preferred site characteristics:

- large, easily accessible opening
- good soil conditions
- 30 plants of 3 or more species
- free from chemical contaminants

Sampling the bioindicator site:

- identify starting point (put on map)
- select plants in a random manner
- do not skip plants with little or no injury
- avoid suppressed or shaded plants
- evaluate leaves on each plant for amount and severity of injury

Ozone injury characteristics:

- on broad-leaf plants injury is present on mid-aged and older leaves on the upper leaf surfaces
- overlapped leaves will have no injury on the bottom leaf
- stippled lesions are uniform in size and shape, most often tiny purple-red to black spots located between the veins on the upper-leaf surface
- on pine, ozone injury is usually present on older needles as small patches of yellow tissue with diffuse borders, surrounded by green tissue
- collect, press, and mail injured leaf samples for injury validation

Injury Scale

Percent Scale for Injury Amount: Estimate and record the percentage of leaves (or needles) on the plant with ozone injury symptoms relative to the total number of leaves (or needles) on the plant.

OZONE II	jury symptoms relative to the total number of leaves (of fleedles) on the plant.
CODE	<u>DEFINITION</u>
0	No injury; the plant does not have any leaves or needles with ozone symptoms.
1	1 to 6 percent of the leaves have ozone symptoms.
2	7 to 25 percent of the leaves are injured.
3	26 to 50 percent of the leaves are injured.
4	51 to 75 percent of the leaves are injured.
5	>75 percent of the leaves have ozone symptoms.

Percent Scale for Severity of Injury: Estimate and record the mean severity of symptoms on injured foliage.

Percent	Scale for Severity of Injury . Estimate and record the mean seventy of symptoms of injured foliage.
CODE	<u>DEFINITION</u>
0	No injury; the plant does not have any leaves or needles with ozone symptoms.
1	On average, 1 to 6 percent of the leaf area of injured leaves have ozone symptoms.
2	On average, 7 to 25 percent of the leaf area of injured leaves have ozone symptoms.
3	On average, 26 to 50 percent of the leaf area of injured leaves have ozone symptoms.
4	On average, 51 to 75 percent of the leaf area of injured leaves have ozone symptoms.
5	On average, >75 percent of the leaf area of injured leaves have ozone symptoms.

Species Codes

Note: (1) The best biomonitoring site is a large opening or stand with <40 percent crown closure where many individuals (>30) of more than 2 species are growing under good conditions of soil nutrition and moisture. The site must be easy to access. (2) A leaf sample must be collected and mailed to the regional ozone expert for every species showing ozone injury symptoms on each site visited by the field crew. Failing to collect a leaf voucher means the data cannot be used.

Web Site address: fiaozone.net

OZONE BIOINDICATOR PLANTS

Data Sheet for the Voucher Leaf Samples

To be filled out by the FIELD CREW or Cooperator: Refer to the Field Guide for code definitions.

State	County	Ozone Hexagon Number	O3Plot Number ¹	Month	Day	Year	Crew ID	Crew Type
								regular OA
								ŲA

¹O3Plot Number refers to the number of locations (1 or 2) used for each hexagon number. Separate sheets should be used for each location.

Crew Name(s)	e-mail address	Phone number

Fill in the required codes. Code definitions are in the Field Guide. For quick reference, see below.

			The first date of the first state		
Bioindicator Species Code Injury Injury		Injury	Is the leaf sample injury close to 100% ozone stipple ($$), or is		
or Common Name	Location	Type	some other upper-leaf-surface injury also present (e.g., insect or fungal lesion)?		
1 st			Close to 100% Estimated percent other		
2 nd			Close to 100% Estimated percent other		
3 rd			Close to 100% Estimated percent other		
4 th			Close to 100% Estimated percent other		

<u>List species</u> evaluated at this location that did not have ozone symptoms: →

Notes: Add notes on the leaf samples, plot conditions, safety, and weather as needed.

122 Ponderosa pine	116 Jeffrey pine	746 Quaking aspen	924 Scouler's willow	351 Red alder
960 Blue elderberry	961 Red elderberry	965 Huckleberry	905 Ninebark	906 Pacific ninebark
907 Western wormwood	908 Mugwort	909 Skunk bush	968 Evening primrose	969 Snowberry

Injury Location Codes:

1= greater than 50% of the injured leaves are younger leaves (broadleaf) or current whorl (pine)

2= greater than 50% of the injured leaves are mid-aged or older (broadleaf) or on whorls 1 year and older (pine)

3= injured leaves are all ages.

Injury Type Codes:

1= greater than 50% of the injury is upper-leaf-surface stipple (broadleaf) or chlorotic mottle (pine).

2= greater than 50% is not stipple (tan flecks, bifacial or general discoloration), or something other than chlorotic mottle (pine)

3= injury is varied or difficult to describe.

Mail this sheet with the leaf samples to:

[Do not write below this line] Pat Temple

USDA FS, PSW Experiment Station

4955 canyon Crest Drive Riverside, CA 92507

QA/QC PERSON: To be filled out by the National Ozone Advisor or Regional Expert. V

-							
	Date checked	Date rechecked	S	Plot Status			
			GOOD Easy to read – ID obvious	FAIR	POOR Samples unreadable or not labeled correctly	(+ozone)	(-ozone)

Bioindicator Species	Positive for ozone	Negative for ozone	Explanation

Additional questions for the data collector: