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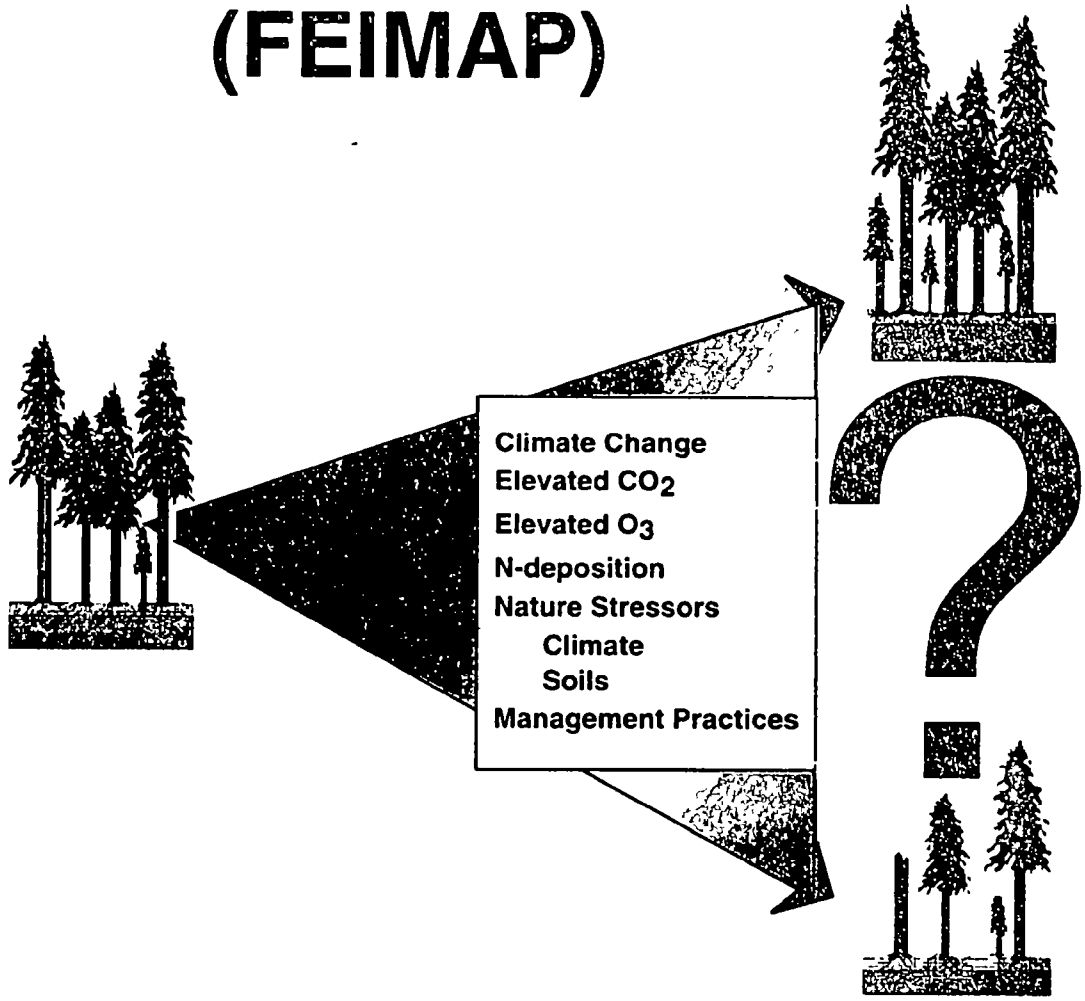
Forest Ecosystem Indicators: Monitoring, Assessment, Prediction (FEIMAP)



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Forest Ecosystem Indicators: Monitoring, Assessment, Prediction (FEIMAP)

by

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Abstract

Ecological indicators for forests are necessary for (1) predicting and/ or assessing the future response of forests to anthropogenic stressors, (2) detecting and quantifying changes and trends in forest condition, (3) linking changes in condition to likely stressors, and (4) identifying early warning measures for loss of integrity and sustainability of ecological resources. The approach for developing indicators includes: (1) process-based models, (2) ecological measures, (3) stress-response data relationships, and methods, at scales from ranging from populations to landscapes. The approach includes collection of climatic, edaphic and ecological data from intensive field sites, spatial data bases on land cover/land use and elevation and controlled chamber experiments to build and parameterize site (e.g. biogeochemical cycling and GAP) and landscape models (e.g. C, N, and water quantity).

Initially, date will be collected along an elevation gradient in the western Cascades of Oregon that will be used to parameterize the MBL-General Ecosystem Model (GEM). This model will be used to assess how changes e.g., in temperature, nitrogen deposition, CO_2 concentration, and soil moisture affect biomass production and ecosystem storage and cycling of C and N, and to identify key ecosystem processes that are sensitive to these stressors. Key processes sensitive to climatic and atmospheric stressors are those related to C and N cycling in the rhizosphere or plant/soil interface and to C and N allocation and partitioning. Therefore, we will develop and evaluate indicators for rhizosphere processes (e.g. structure and function soil food web), ecophysiological processes (e.g. carbon allocation to fine roots), and ecosystem and landscape function. We will use other sites in the western Cascade region and Olympic National Park to validate and verify the performance of the models and potential indicators.

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1. INTRODUCTION

1.1. Research Needs within EPA Office of Research and Development

The US Environmental Protection Agency (EPA) is required to protect the environment to ensure clean air and water and an uncontaminated food supply. Although many environmental problems have diminished as a result of such Federal actions, some have not and new problems continue to arise. To deal with the new and potential problems, the EPA has established ecological protection as one of its highest priority research areas.

The EPA's Office of Research and Development (ORD) has substantially changed its organization and operation to strengthen the Agency's science base and improve the nation's ability to effectively respond to complex environmental challenges (US EPA 1996) One of the most important changes is the explicit use of the risk assessment and management paradigms to shape and focus the Agency's research agenda. One of the key research needs for improving ecological risk management is to provide the scientific tools necessary for cost-effective management decisions by stakeholders on the protection of ecological resources at local, regional, and national scales. Such scientific tools need to: identify the most important ecological risks; perform risk assessments, including problem formulation, characterize stress-response and exposure functions, and characterize risks; identify and evaluate risk management options; and verify and monitor measures or indicators of risk reduction (US EPA 1996).

The Committee on Environment and Natural Resources (CENR) calls for all environmental agencies to merge efforts in forming a national monitoring and research network to achieve the national common goal of understanding and managing ecological systems for their continued and sustained vitality, diversity, habitat, and ability to provide goods, services, and enjoyment for humans. In response to this national goal, EPA/ORD focuses its research efforts through the Environmental Monitoring and Assessment Program (EMAP) to: (1) develop ecological indicators that can be used to monitor status and trends in ecosystems, (2) design effective systems for monitoring status and trends, and (3) integrate and synthesize environmental data (US EPA 1997a).

Here we present a plan for a long-term research project to support several of the Agency's research needs with reference to forests. Specifically, the overall goal of this plan is to develop and evaluate ecological indicators that support (1) EMAP's need for forest indicators of ecological integrity and sustainability (US EPA 1997c), (2) the Global Change program's need for indicators of global change, and (3) risk assessment and risk management protocols. We consider ecological indicators to include predictive models to identify potential risks to global change and air pollutants, stress-response functions, methods, and measures and their behavior over time and space. These indicators can be used to

assess and monitor change and trends in the condition of forest resources, link changes to likely stressors, predict response of forests to future environmental change, and identify early warning signals.

1.2. Current Status on Forest Indicators for Assessment and Monitoring

In 1989, the EPA in collaboration with other federal and state agencies and research institutions, initiated the Environmental Monitoring and Assessment Program (EMAP). The EMAP is a national program designed to assess the status and trends of the Nation's ecological resources, including forests, arid lands, agroecosystems, wetlands, inland surface waters, and estuaries (Hunsaker and Carpenter 1990). The program was initiated as a long-term, interagency project to monitor and evaluate the condition of ecological resources, develop methods for anticipating emerging problems before they reach crisis proportions, and contribute information to decisions on environmental protection and management at regional and national scales (Thornton et al. 1993).

At present, surveys of forests condition are conducted by USDA Forest Service through the Forest Health Monitoring (FHM) program. The FHM program began in 1990 in the Northeastern region of the USA in partnership with EMAP. The FHM program has four components: detection monitoring, evaluation monitoring, intensive site monitoring, and research on monitoring techniques. Detection monitoring is the most developed component of the program. It is composed of a nation-wide network of long-term monitoring plots that have been or are being established throughout most of the forested areas of the US. The FHM program also uses information from other programs both within the Forest Service (e.g., Forest Inventory and Analysis, fire data, and forest pests and disease data) and other agencies (e.g., EPA for air pollution data and NOAA for weather data). A suite of indicators, considered important in assessing forest health, have been developed (Lewis and Conkling n.d.) and are being monitored on the network of FHM plots. Four groups of indicators are being used (A. J. R. Gillespie, USFS, Radnor, PA, pers. comm.): mensuration (e.g., tree growth, mortality, dendrochronology), crown condition (e.g., LAI, crown dieback, and branch evaluation), tree damage (e g., leaf coloration and stem damage), and ozone sensitive plants (e.g., lichens and bioindicator plants). The FHM program uses these indicators to evaluate the current forest condition and to determine if forest health is static, improving, or degrading over time.

Within the Pacific Northwest forests, a FHM pilot study was established in 1994 (Campbell and Liegel 1996) using 25 1-ha sample plots located in forests of the Cascade Range of Oregon and Washington. Within these plots cover and species composition of understory vegetation, species composition and structure (e.g., size classes, tree density, basal area, number of large live and dead trees) of the overstory vegetation, crown condition (e.g., vigor, density, dieback), tree damage by type (e.g., cankers, wounds, damaged foliage), species richness of macro-lichens on woody substrates, and abundance of

songbirds in relation to forest habitat are being measured (Campbell and Liegel 1996) Results from this pilot study have provided limited baseline data on the condition of the forests in the Cascades as well as testing the FHM sampling protocols for the region.

Several scientific concerns and challenges have emerged during reviews of the EMAP by the EPA Scientific Advisory Board, the National Research Council (NRC 1995), EPA/ORD, and from recent research findings at the EPA Western Ecology Division (WED) of NHEERL relative to forest indicator development (Table 1). The NRC (1995) concluded that a focused research program on indicator development is needed because the use of scientifically defensible indicators should be the heart of the EMAP.

The development of ecological indicators for forests represents a unique challenge. For example, forests are dominated by long-lived species, have simple to complex species assemblages, contain multiple vertical layers, composed of single- or mixed-age individuals, grow in complex terrains and environments, depend primarily upon nutrient recycling rather than external inputs, are limited by a myriad of environmental factors that change throughout the lifetime of the forest, and are subject to an array of natural (e.g., fire, pests) and anthropogenic (e.g., climate change, N deposition, tropospheric ozone, UVB, management, chemical application, exotic invasions) stressors of different intensities and duration (Aber and Melillo 1991). Further, the effects of various anthropogenic stressors are likely to differ depending upon the ecological conditions under which forests grow For example, ozone effects in arid regions are different than in humid ones, and increased N deposition is likely to affect N limited forests differently than N saturated forests.

Despite the concerns that have emerged from reviews of EMAP (Table 1), the present suite of EMAP/FHM indicators are useful for monitoring the status of forests. However, they provide a signal late in the response phase given the longevity of trees; indicators are needed that occur early enough during response to stress that action can be taken. We believe that indicators of this nature will be those that are more direct measures of forest processes or those structural components that have fast turnovers, such as plant resource utilization, fine root dynamics, and the soil food web. The reason for focusing on forests processes or components with fast turnover is that they are more sensitive to change, i.e., they generally have faster response times whereas major structural components have slow turnover and response times (decades or more).

Table 1. Scientific concerns and challenges that have emerged from reviews of the EMAP

- Current indicators place heavy reliance on an epidemiological model which lacks the predictive power to determine how changes in ecosystem properties (e.g., nutrient cycling, nutrient loss, productivity, or biodiversity) will respond to stress.
- Current indicators are not sufficient to link changes to likely stressors nor identify early warning signals.
- Current indicators cannot predict the future status of ecosystems under changed conditions.
- Current indicators do not include measures of the plant/soil interface, which is critical in maintaining ecosystem structure and function (Andersen and Rygiewicz 1996, Rygiewicz and Ingham in press).
- Current indicators do not account for a plant's ability to survive environmental stresses by adjusting processes such as carbon allocation and partitioning, nutrient uptake, and water use (Hogsett et al. 1985a, 1988 Andersen et al. 1991, 1997b).
- Current indicator development has not given priority to measurements that integrate limiting factors (e.g., nutrient availability) over the growing season.
- EMAP has not developed an indicator program that adequately separates changes induced by stress from natural cycles of disturbance from anthropogenic sources.

2. RESEARCH APPROACH

2.1. Research Objectives

The goal of this research project is to develop, test, and evaluate ecological indicators necessary to (1) predict or assess future response of forest ecosystems to anthropogenic stressors, (2) detect and quantify status and trends in forest ecosystem condition, (3) link changes in condition to likely stressors, and (4) identify early warning measures for loss of integrity and sustainability of forest ecosystems in the Pacific Northwest.

By ecological indicator we mean a measurement or group of measurements that can be used to describe or predict the condition and change of an ecosystem over time, or of one or more of its critical processes or components over time. Thus indicators will include ecological measures, stress-response functions, methods, and process-based models, at scales from populations to landscapes. Examples of ecological indicators include stress-response functions and predictions based on simple regression models to more complex process-based ecosystem or landscape models; or they might include simple measures of an ecological process, surrogate measurements of an ecological

process, and/or combinations of measurements of key processes or components.

2.2. General Approach

The research will focus on developing ecological indicators that build on the expertise of scientists at NHEERL-WED, which includes: (1) process-based modeling and extrapolation to landscape scales and (2) experimental work on anthropogenic stress-effects (e.g., ozone, and elevated CO_2 concentrations and temperature) on forest species, including C and N allocation and partitioning, root dynamics, and composition and function of the soil food web. Results from previous and ongoing research at WED suggest four potentially fruitful areas of investigation for developing ecological indicators: (1) modeling biogeochemical cycles and changing structure of forest stands (Solomon and Leemans 1990, Solomon and Bartlein 1992, McKane et al. 1997a,b), (2) modeling and analyzing relations between ecological processes and landscape pattern (Cramer and Solomon 1993, Solomon and Shugart 1993, Brown et al. 1993, 1994, Brown and Gaston 1995), (3) measuring processes of resource utilization by producers (Andersen et al. 1991, 1997b; Hogsett et al. 1985, 1988; Tingey et al. 1996, 1997), and (4) measuring resource utilization by consumers, including soil food web structure and function (Andersen and Rygiewicz 1995; Porteous et al. 1994, Donegan et al. 1995, Seidler and Fredrickson 1995, Widmer et al. 1997a).

Through the combined use of field data, remotely-sensed and other spatial data bases, and simulation modeling we will produce measures, methods, stress-response functions, and models from which indicators will be developed and evaluated The ecological indicators will be derived from various ecosystem components and processes and represent a range of scales from populations, to ecosystems, and to the landscape level (Figure 2-1).

Two types of process-based models will be included in this research: (1) biogeochemical models with an emphasis on C and N cycling and (2) forest succession models to address changes in stand structure (tree species composition and age-size distributions) with succession (Fig. 2-1). These models are driven by a similar set of environmental factors and will be used to identify key sensitive ecosystem processes and components as well as to assess and predict the effects of changes in temperature and precipitation, elevated atmospheric CO_2 concentrations, air pollutants, N deposition, forest management, and natural disturbance regimes on Pacific Northwest forest ecosystems.

Landscapes encompass a variety of climatic, edaphic, and geomorphologic factors all of which influence how ecosystems respond to an individual or suite of anthropogenic stressors. By coupling site models with spatial data bases of the biophysical factors, we plan to investigate how anthropogenic stressors, singly and in combination, affect the functioning of landscapes (Fig. 2-1). In particular, we plan to investigate how different patterns of land use affect ecological processes such as landscape-level primary productivity, N losses, and water quality. Furthermore, we also plan to

investigate how other stressors such as N deposition and climate change interact with changes in land use to affect landscape-level processes previously mentioned.

The field data collection phase will serve three roles: (1) to parameterize, calibrate, modify, and test the simulation models to be used in this research, (2) to determine the natural variation over time and space of ecological processes that could serve as indicators, and (3) to elucidate the mechanisms that are involved in key ecological processes. Similar measurements of the same processes will also be collected in on-site experimental facilities to allow us to determine the response of the same ecological processes under known stressors.

Promising indicators for wider -scale adoption for forest monitoring selected from the research activities will be those that have low spatial and temporal variation and maintain a strong correlation with changes in natural stress

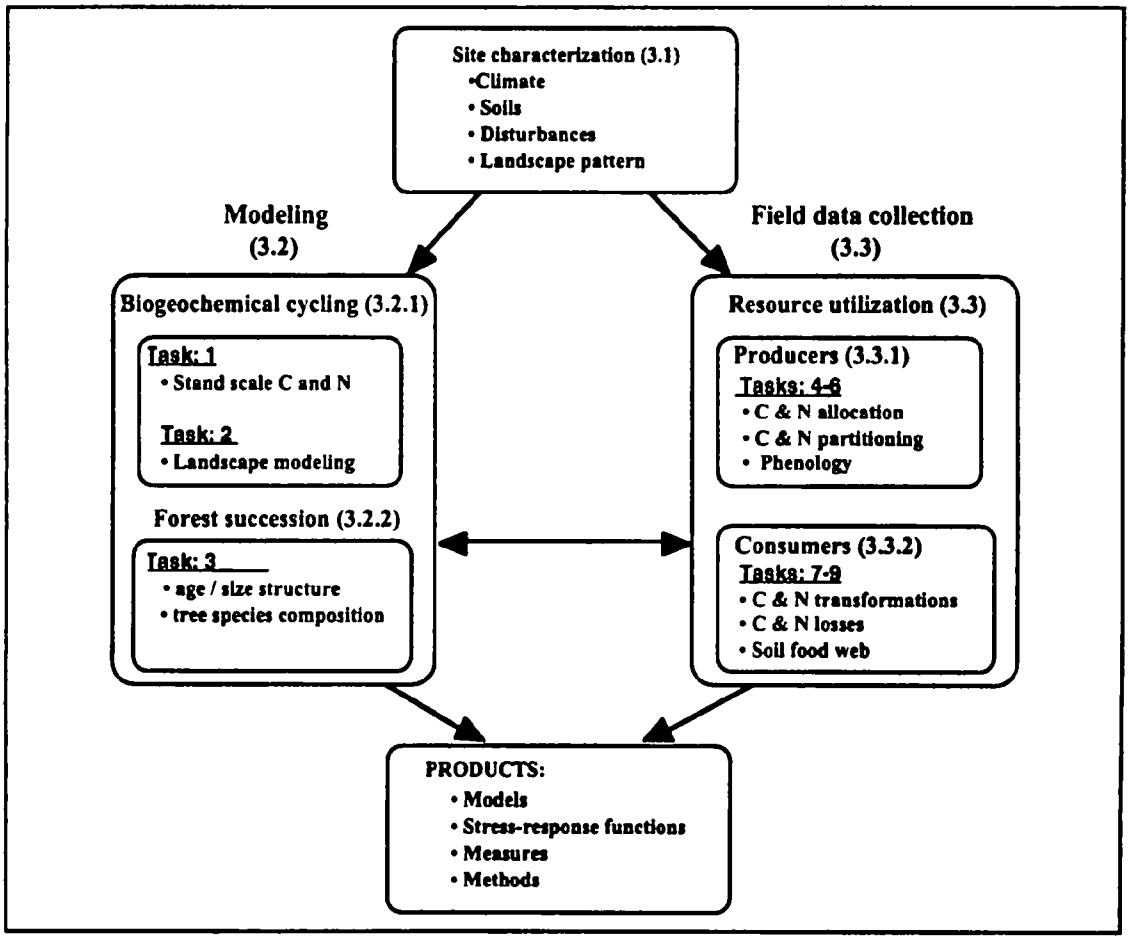

Figure 2-1. Linkages among the various phases of the research plan and the outputs suitable for assessing and monitoring forests. The task numbers and numbers in parentheses refer to the sectional numbering scheme used in Section 3 where details for each component of the research are given.

factors in these complex environments. These promising indicators will then be subjected to sensitivity analyses. Here, extensive field sites across climatic gradients and long-term data from other established research sites in representative forests of the western Cascades will be used to test the sensitivity of indicators to changes in climate, land use/land management, and pollutants. Finally, indicators proven useful in specific areas will be evaluated for regional application, e.g., throughout forests of the Pacific Northwest. Modeling activities will focus on validation to see if predictions hold across the range of response and environmental conditions, and ultimately to evaluate indicator sensitivity. Indicator selection will be based on comparative sensitivity analyses across sites and conditions and on EMAP selection criteria and guidelines (US EPA 1997a).

3. RESEARCH IMPLEMENTATION

The research will focus on developing indicators for forest ecosystems. Most of the research described here will be done in field sites to determine the natural variation over time and space of ecological processes. This section contains a description of the research field sites (3.1), including data collection for characterizing the sites. In this Project, several potential types of indicators (Fig. 2-1) will be evaluated and/or developed. They include:

- Modeling biogeochemical cycling at the stand and landscape scale (3.2.1)
- Modeling forest succession processes at stand to landscape scales (3.2.2)
- Measuring resource utilization (C and N) by producers (3.3.1)
- Measuring resource utilization (C and N) by consumers (3.3.2)

Similar modeling approaches and measures of resource utilization have been or are being investigated in three other projects at WED: *Effects of CO_2 and Climate Change on Forest Trees* (TERA I experiment), *Interactive Effects of O_3 and CO_2 on the Ponderosa Pine Plant/Litter/Soil System* (TERA II experiment), *Effects of tropospheric ozone on forest trees* (Forest Ozone project) (see Section 4. for further details on linkages with other research programs in WED).

3.1. Field Sites

The research activities described here will take place in intensive field sites, extensive field sites, and whole watersheds. Here we describe the field sites, and types of data that have been or will be collected to characterize these study sites (Fig. 2-1). These data will be used to interpret trends in field data collection as well as to drive the site and landscape models.

3.1.1. Intensive field sites

The intensive sites were established to provide an understanding of ecosystem and ecophysiological processes (e.g., nutrient fluxes, net primary production) and to provide data for model parameterizations. This understanding is achieved by having a number of investigators conduct a range of studies and insuring that a common set of environmental data is collected and made available to all.

The ecological processes and forest composition in the western Cascades varies in response to several factors including: climatic gradients and variability, soil type, management activities, successional stage, disturbances (e.g., fire and insects), and intersite and interannual changes. To capture the variability associated with these factors, we have established and will establish additional intensive field sites in the Western Cascades. We will measure a suite of parameters at our intensive sites for a minimum of 5 yr to capture the interannual variation in ecological processes resulting from climatic variability.

Our proposed indicators research project will take advantage of established sites when possible (Appendix B). For example, existing research sites established at the H. J. Andrews Experimental Forest offer the advantages of long-term data bases on forest ecosystem response. However, we believe it is also important to establish a core set of sites that can be measured and/or manipulated strictly for the purposes of the research proposed here. There are two main reasons for this:

1) To be of optimum use as for our project, most proposed measures must be linked to a data set that describes all of the major pools and fluxes of C and N in the ecosystem, as well as all of the environmental conditions that may have a bearing on the response of a measure. To our knowledge, no other existing sites outside of our current intensive research sites meet this need. For example, there are sites in Oregon and Washington where research projects are already underway to describe ecosystem C budgets in detail (e.g., OTTER and Wind River Canopy Crane), however, complimentary data on N budgets are being given less emphasis. Because of the importance of N in limiting forest growth, it is essential that C and N budgets be described together for the same sites, not inferred from other nearby sites that may or may not have similar soils, vegetation or climate.

2) Activity or "wear and tear" associated with intensive measurements at a research site can have adverse effects on ecosystem processes. For example, foot traffic can disrupt growth of fungal hyphae and fine roots or alter rates of net N mineralization. Thus, a major challenge in conducting our research is to avoid the Heisenberg Principle, whereby the act of measurement alters the very thing that is to be measured. By establishing sites that we are responsible for managing, it will be logistically easier to regulate where foot traffic is allowed and what areas are available for different research activities. This becomes very important in providing researchers with a history of what activities have taken place in any given location, so that potential problems can be avoided. Our goal will be to facilitate collection of high quality data while providing for the long-term maintenance of the research sites.

Current field sites

Our current research sites include both low and high elevation forests and clearcuts located in the western Cascades along the Highway 20 corridor between Sweet Home, OR and the Santiam Pass (Fig. 3-1). These sites differ in climate (e.g., growing, temperature, precipitation form and amount) and soil nutrient levels. In this area many disturbances typical of the western Cascades are at work including flooding, insect and disease outbreaks, and wind storms. However, fire and human impacts (including logging and road building) are the dominant disturbances that have affected the conditions within these forests. Historic fire return intervals vary from 25 to 110 years for low-intensity ground fires, to 100-200 years and more for high-intensity stand-replacement fires (Morrison and Swanson 1990).

The Falls Creek Site is located at an elevation of 537 m in the western hemlock (*Tsuga heteropyhlla*) vegetation zone (Franklin and Dyrness 1988) in the South Santiam River drainage. The development of dense, essentially even-aged stands of Douglas fir is a common occurrence after a wildfire or logging. These stands can be dense enough to delay the establishment of understory vegetation. Re-establishment of the characteristic understory and invasion of western hemlock occurs as mortality begins to open up the overstory (Franklin and Dyrness, 1988). Historic records reveal a large stand-replacement fire (Moose Mountain fire) in 1856 followed by a 100 year storm in 1861. However, tree-ring analyses show that the oldest Douglas firs are 100 to 110 yr old. In 1969, there was a commercial thinning. About 16 ha was clear-cut at the Falls Creek Site in 1988, broadcast burned in 1990, and replanted

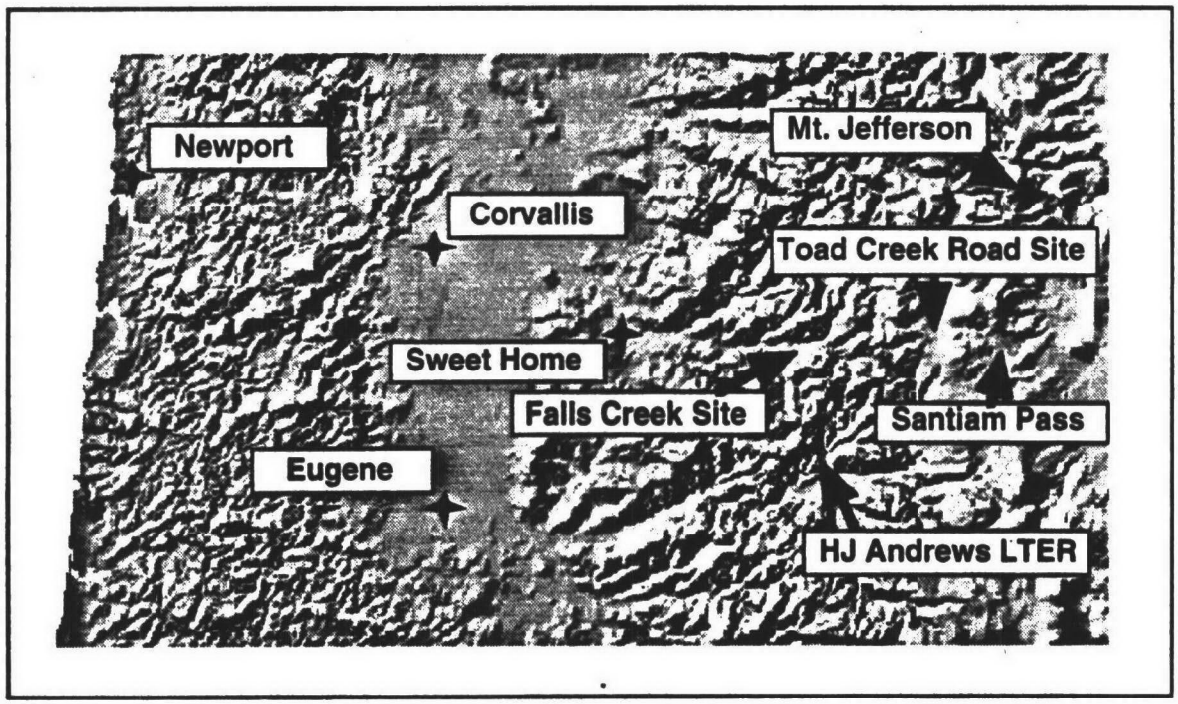

Figure 3-1. Topographic map showing the locations of the current research sites in relation to other points of interest.

to Douglas fir in 1993. The area is currently managed as elk wintering and calving habitat; consequently, the area was aerially seeded with legumes and grasses. To maintain the forage, most of the clear-cut, except where we are conducting our studies, was fertilized (spring, 1994 and 1996).

We established research plots in both the clear cut and the adjacent 140-year-old forest in 1995. Douglas fir is the dominant overstory tree with western hemlock and vine maple occurring in the understory. The current stand density for trees >20 cm dbh is approximately 238 trees/ha. In 1995 we established a 75 m x 90 m study area in the forest in which we sampled 12 randomly located 15 m x 15 m plots. Trees range in diameter from 2 to 90 cm and up to 60 m tall (Table 3-1). A vegetation survey in 1996 found a total of 16 plant genera (not including tree species), with a mean percent cover in m^2 forest plots of 49%.

Vegetation studies in the clear cut in 1996 indicated the presence of at least 16 plant genera, including the four genera that were aerially seeded in 1990. Mean percent cover in the low site m^2 clearcut plots was 99%.

Table 3-1. The composition and typical sizes of the trees on Falls Creek and Toad Creek Road Site forest plots.

Plant species	Mean and max. height (m)	Mean and max. diameter (cm)
Falls Creek Site		
Douglas fir	49, 64	61, 91
Western hemlock	4, 36	4, 27
Toad Creek Road Site		
Douglas fir	44, 61	74, 111
Western hemlock	7, 45	12, 76
Pacific silver fir	7, 29	12, 46

The Falls Creek Site is located on deep to very deep well-drained soils derived from colluvium, glacial till, and alluvium (Legard and Meyer 1973). Bedrock materials consist of andesites, basalt, tuffs and breccias. This soil type is typically found between about 460 and 1370 m. A tentative classification of this soil is coarse-loamy, mesic, Typic Hapludand (Appendix A).

The western hemlock vegetation zone has a wet, mild maritime climate (Franklin and Dyrness 1988). As the Low Site lies on the eastern side of this zone some distance from the ocean, there can be large seasonal variations in soil moisture and temperature.

Precipitation occurs mainly during the winter; summers are relatively dry and account for only 6 to 9% of the total precipitation (Franklin and Dyrness 1988). Meteorological stations were established in the clear cut in 1994 and in the forest in 1995. Table 3-2 shows the types of environmental data collected in the clear cut and forest locations and Table 3-3 gives typical values for 1995.

Table 3-2. Types of meteorological and soils data collected at the Falls Creek and Toad Creek Road Sites in both the clear cuts and late successional forests.

Parameters*	Falls Creek Site		Toad Creek Road Site	
	Clear-cut	Mature forest	Clear-cut	Mature forest
Air temperature - C	√	√	√	√
Relative Humidity - %	√	√	√	√
Solar Radiation - PAR	√	√	√	√
Solar Radiation - Total radiation		√		√
Precipitation - mm/hr	√	√	√	√
Wind speed - m/sec	√	√	√	√
Soil temperature - C (several depths)	√	√	√	√
Soil moisture - % (TDR)	√	√	√	√
Soil moisture - % (Reflectometer)	√	√	√	√
Snow depth			√	√

*Data are collected as 1-hour averages except for soil moisture which is collected as 3-hour averages or periodically depending on the measurement system.

Table 3-3. Typical climate conditions measured in the clear cut at the Falls Creek and Toad Creek Road Sites for the calendar year 1995.

Mean annual air temperature (°C)	Mean annual relative humidity (%)	Mean annual daily total PAR (µmol m^{-2} day^{-1})	Total annual precipitation (mm)	Growing degree-days	Maximum snow depth (cm)
Falls Creek Site					
10.5	82	7246	1798	3656	NA
Toad Creek Road Site					
7.6	76	6895	1947	2673	127

The Toad Creek Road Site is located at an elevation of 1220 m in the Pacific silver fir (*Abies amabalis*) vegetation zone (Franklin and Dyrness 1988) in the upper reaches of the McKenzie River drainage. In this vegetation zone, a typical successional sequence begins with the establishment of Douglas fir and/or noble fir (*Abies procera*), followed by the shade tolerant western hemlock and Pacific silver fir which typically develop later under a forest canopy (Franklin and Dyrness 1988). Up to 500 years following a disturbance, a typical mixed stand includes scattered, large Douglas fir, abundant but smaller western hemlock, and abundant seedlings, saplings and poles of Pacific silver fir (Franklin and Dyrness, 1988). In the high site forest, 28 plant genera (excluding the trees species) were identified in 1996, with a mean percent cover in m^2 plots of 78%.

Douglas-fir is the dominant tree species in the Toad Creek Road Site forest with western hemlock and Pacific silver fir occurring in the understory. The dominant Douglas firs are between 200-220 yr old based on tree-ring counts. The even-aged structure of the dominant Douglas-fir in this forest is consistent with typical patterns of natural regeneration following high-intensity stand-replacement fires. Current stand density of trees >20 cm dbh is approximately 267 trees/ha. This forest is classed as late successional/old growth. In 1995 we established a 75m x 75 m area in the forest in which we sampled 11 randomly located 15 m x 15 m plots. The composition and sizes of the trees on these plots is given in Table 3-1.

Following the 1991 clear cut of about 18 ha, the site was replanted in 1994 with a mixture of Douglas-fir (15.3%) noble fir (73.1%), grand fir (*Abies grandis*) (7.5%) and western white pine *(Pinus monticola)* (4.2%) at a density of 212 trees/ha. The site was not burned following the clear cut and most of the large woody debris was left on the site. In 1996, a vegetation survey found 28 plant genera with a mean percent cover in m^2 plots of 42%.

The Toad Creek Road Site is located on soils derived from volcanic ejecta and glacial till overlying bedrock of hard andesites and basalt (Legard and Meyer 1973). This soil type occurs at elevations of 850 to 1250 m on glacially smoothed lava flows. In general, the soil has a fine loam to loam texture with a medium granular structure. The soil has been classified as a coarse-loamy, mixed, frigid, Typic Hapludand (Appendix A).

The Pacific silver fir vegetation zone is wetter and cooler than the adjacent western hemlock vegetation zone and receives considerably more precipitation in the form of snow. Winter snow packs of 1 to 3 meters are common (Franklin and Dyrness 1988). Meteorological stations were established in the clear-cut in 1994 and in the forest in 1995. The types of environmental data collected and typical climate conditions at the High Site are shown in Tables 3-2 and 3-3, respectively.

3.1.2. New Research Sites

To accomplish the scientific objectives of the Project, we will establish additional research sites. These sites will be selected to: (1) meet one or more of the site selection criteria listed below, (2) maximize

the opportunities for multiple investigators to use the same sites and (3) be located adjacent to or near a meterological station operated by the Project or other group (e.g., SNOTEL).

Criteria for site selection: (1) provide replication of the existing Falls Creek and Toad Creek Road sites, (2) test specific hypotheses and (3) provide data for model development or testing.

Site Replication - A major constraint of most ecological research is the lack of replicated field sites. This lack makes difficult the interpretation of variation observed at the unreplicated sites. We propose to establish new research sites to serve as replicates of the existing sites (Falls Creek and Toad Creek Road) and also establish the variation in ecological processes due to the intersite variability. The replicates are Moose Mountain for Falls Creek and Soapgrass Mountain for Toad Creek Road. The Moose Mountain site is at approximately the same elevation and contains the same general vegetation as the Falls Creek site (Douglas Fir, Vine Maple, Oregon Grape and Salal). The land type is also the same between the sites but Moose Mountain has a southern exposure. A battery of soil analyses will be obtained to compare physical and chemical composition with that of the existing Falls Creek site. The Soapgrass Mountain site also has a similar elevation, climate and vegetation type as the Toad Creek site. Initial measurements suggest that it may be a wetter site with a high soil nitrogen level. If detailed measures confirm the initial observations, these differences will be accounted for in comparing sites.

Test Specific Hypotheses - Several ecophysiological processes are outlined in tasks 4 through 6 for indicator development. These studies are appropriately addressed by testing specific hypotheses. To achieve the conditons for testing may require additional sites or the use of existing sites.

For example, previous studies have established baseline biological and nitrogen fixation data for the Falls Creek site. A new experimental site at Moose Mountain has recently been established to carry out intensive soil biological analyses and nitrogen fixation studies to compare with the existing data from Falls Creek. Also the Moose Mountain site will be used to establish a data set prior to, immediately following and for about 5 years from when the clearcut is established in the summer of 1998. Replanting with Douglas Fir will take place within 12 months after the clearcut is established. This unique management scenario will allow us to follow changes in the food web biota and nitrogen fixation over a time sequence of a major management perturbation

Within the Moose Mountain study area there is a 41 acre clear cut (designated as powder Regen III) scheduled for the summer of 1998. A forested site, several hundred feet from the clear cut on a similar south facing slope, as has been identified as a control. Meteorological stations will be located in both the forested and clear cut sites to monitor soil and litter temperature and moisture, air temperature, precipitation and wind speed. Thus, comparison of the kinds of diversity of biological endpoints with the Falls Creek site can be linked to similarities or differences in the

meteorological conditions as well as to major management disturbances e.g. the clear cut (see Subtask 9b for a detailed explanation of the endpoints to be examined).

A second example of site selection to test specific hypotheses involves Toad Creek Road and Soapgrass Mountain. The hypothesis is that N retranslocation is influenced by soil N levels. These two sites were selected because that have similar elevation, climate and vegetation characteristics but differ in soil N. Total soil N decreases 0.4% at Soapgrass to 0.1% at Toad Creek Road.

Model Development - Parameterization, calibration and testing of biogeochemical and stand models requires a range of data from a number of different sites. The existing intensive sites (Falls Creek and Toad Creek Road) have been used to develop a parameter set for the biogeochemical model. Additional sites will be selected to: (1) establish the maximum biomass which is needed to constrain the model simulations, (2) provide geographically extensive data to extend the current biogeochemical model parameter set to a regional parameter set using data from the Cascade Center for Ecosystem Management Permanent Study Plots and (3) test the accuracy and reliability of the regional biogeochemical model parameter set. The model will be tested using new data from a range of sites in the Olympic National Park representing a range of vegetation types, precipitation ranges and elevations.

Additional Climate Stations - To provide climate and soil data, we will also establish additional weather monitoring sites concurrent with the establishment of the new research sites. In addition to our monitoring sites, the US Department of Agriculture, Natural Resources Conservation Service maintains a number of SNOTEL sites within the Santiam River Basin and adjacent drainages where daily temperature (maximum, minimum and mean), precipitation, and snow water equivalents are measured. If we locate some of our new plots adjacent to the SNOTEL sites we will use their data rather than establishing our own sites.

3.1.3. Watersheds

Abundant spatial data bases on subwatershed boundaries, seral stage, climatic, soils, terrain (digital elevation models [DEMs] of 30 m to 500 m resolution), ownership, harvest history, and vegetation are available for most of the Pacific Northwest region. These data bases include products derived from remotely-sensed imagery ranging from fine-scale aerial photography and Landsat TM satellite imagery with spatial resolutions of 30 m or less, up to more coarse scale AVHRR imagery at resolutions of 1.1 km. For example, Cohen et al. (1996) have used Landsat TM imagery to map six forest successional stages and harvest incidence between 1972 and 1991 in a 1.2×10^6 ha central Oregon Cascades landscape.

We propose to work at the watershed scale initially within the 5.1×10^4 ha South Santiam River Basin. This is also where our Low Site is located (Section 3.1.1). Situated in one of the major drainages of the western Oregon Cascades, the watershed has been described by ownership, stream systems,

current and historical seral stage, road network, and fire history (Sweet Home Ranger District 1995). The watershed is composed of 10 subwatersheds ranging in size from 1097 to 9630 ha and varying from stand initiation to late-successional/old-growth seral stages. Copies of the digital maps of these data have been obtained. Further, the data of Cohen et al. (1996) have been obtained.

3.2. Modeling

Models play a prominent role in risk assessment because they are the primary means for relating stressors to probable effects, a conceptual basis for integrating diverse measures into a self-consistent framework, and for making meaningful extrapolations across scales of time, space, and biological organization (Suter 1993b, Rapport 1992, Rastetter 1996). The synergistic interactions in nature among the various environmental driving forces, such as temperature, precipitation, nutrient inputs, topography, and soil moisture, make it impossible to predict or assess future response of ecosystems to anthropogenic stressors, such as air pollutants, climate change, and land use, based on single-factor experiments alone (Rastetter et al. 1991). Process-based models such as biogeochemical cycling models or forest succession models can help improve such assessments by providing a self-consistent synthesis of the results of many experiments. The synthesis provided by these models includes the interactions among ecosystem processes that give rise to the synergistic responses to multiple factors.

The problem with models, however, is that their long-term predictions are impossible to test unambiguously except by allowing enough time for the full ecosystem response to develop. Unfortunately, when potentially significant changes to ecosystems must be assessed, time becomes a luxury (Rastetter 1996). Confidence in models, therefore, has to be built through evaluation against corroborating evidence. Such evaluations may include: (1) comparisons against short-term experiments (less than 10 yr), (2) using space for time substitutions, (3) using reconstruction of past responses, and (4) comparisons with other similar model outputs (Rastetter 1996). Although these methods can be used to evaluate models, none can meet the crucial needs for rigorous testing. This does not mean to downplay the importance of models; their advantage is that they can be used to synthesize all the empirical evidence in a self consistent set of interpretations (Rastetter 1996). Thus models are a vital part of any risk assessment of the response of ecosystems to anthropogenic stressors.

Two models will be used in this research plan to integrate experimental and field data for predicting effects of anthropogenic and natural stressors, for developing stress-response data bases, and for determining sensitive processes or components that can serve as potential early warning indicators The two models, a biogeochemical model (Section 3.2.1) and a forest succession model (Section 3.2.2) focus on different aspects of forests, and complement each other as predictive tools. Indeed, the dynamic forest processes which they simulate are entirely different, although several model output variables can be

compared between the two models (e.g., stand biomass, stand leaf area, nutrient content, etc.).

The biogeochemical models focus on the processes of carbon and nutrient utilization (acquisition, allocation, and partitioning) in plants and soils and how utilization patterns are impacted over time by a suite of anthropogenic stressors such as changes in climate or atmospheric composition, and by forest management. In contrast, forest succession models focus on year to year stability and change in forest tree size classes, age classes and species, as controlled by establishment, growth and mortality processes. The models mimic the gradual replacement of fast-growing and shade-intolerant trees by slower-growing and shade tolerant species, generating the time-ordered implications of this successional process to shifts in community-level carbon storage, soil nutrient status, and vulnerability to anthropogenic stressors. The succession models evaluate how forest stand structure and composition are modulated over time by changes in climate, atmospheric chemistry, species availability and management choices.

Both models will be used to simulate forest dynamics at local scales, and at landscape scales by linking each model to additional spatial data bases. We also plan to integrate ("hard wire") the two kinds of models to simulate and evaluate the importance of the feedback processes between biogeochemical and structural/ compositional attributes and processes, although the tasks required to attain this longer range goal cannot be defined until considerable progress has been obtained on the separate model development paths proposed below (Sections 3.2.1, 3.2.2). The two models require unique suites of parameters, and a common set of driving variables. The data collected for site and landscape characterization (Section 3.1) will serve as some of the driving variables for both models. In the following sections, we present the approaches and data needs to parameterize, simulate and evaluate each model at the stand and landscape scale.

3.2.1 Biogeochemical cycling modeling

Task 1: Modeling at the stand scale

Objectives and background

The basic objectives of this task are to parameterize, evaluate, and verify a model of biogeochemical cycling of C and N in a forested ecosystem at the stand scale (Fig. 3-2) for use as an assessment and predictive tool. The large number of ecosystem processes and components that may be affected by anthropogenic stressors constrains the assessment of stressor-effects by experimentation alone. An alternative is to use process-based models to predict how forest ecosystems will respond to existing or projected scenarios of stressors. Specifically, we will use a biogeochemical cycling model to determine the effects of natural and anthropogenic stressors combined with future forest management actions on PNW forests. For example, the model can be used to determine the relative risk of increased temperature or N-deposition on forests under different harvesting scenarios or to assess the likely effects of forests on various actions taken to control

atmospheric CO_2 concentrations. The model also directly supports risk assessment by providing a tool that can be used in the problem formulation and risk characterization steps. The experimental data developed to parameterize the model and the results of the model simulations will provide stress-response functions (i.e., effects data) that are also needed to conduct a risk assessment for PNW forests.

Selection of biogeochemical model

There are a number of models available that simulate biogeochemical cycling in forest ecosystems (Perruchoud and Fischlin 1995), including CASA (Potter et al. 1993), CENTURY (Parton et al. 1987), FOREST-BGC (Running 1994), LINKAGES (Pastor and Post 1986), MBL-GEM (Rastetter et al. 1991), and TEM (Raich et al. 1991). While all of these models provide a process-based view of ecosystem C and N cycling, they differ with respect to model structure (number of plant and soil compartments), the incorporation of particular processes, coupling with the abiotic environment, and method of model calibration. For our objectives, we require a model that can address the responses of forest ecosystems to changes in CO_2, climate, N deposition, and air pollutants, and that these responses will encompass enzymatic controls on C and N acquisition, stoichiometric shifts in tissues, changes in plant biomass allocation among tissues, altered rates of organic matter turnover and N mineralization, and ultimately a redistribution of C and N between vegetation and soils. Because it would be extremely difficult to obtain sufficient fine-scale data to characterize many of these processes, we also require a model that can be calibrated to infer the needed information from data that are more easily obtained, namely, data collected at the scales of ecosystems (e.g., net primary production) and regions (e.g., vegetation C stocks along temperature and precipitation gradients).

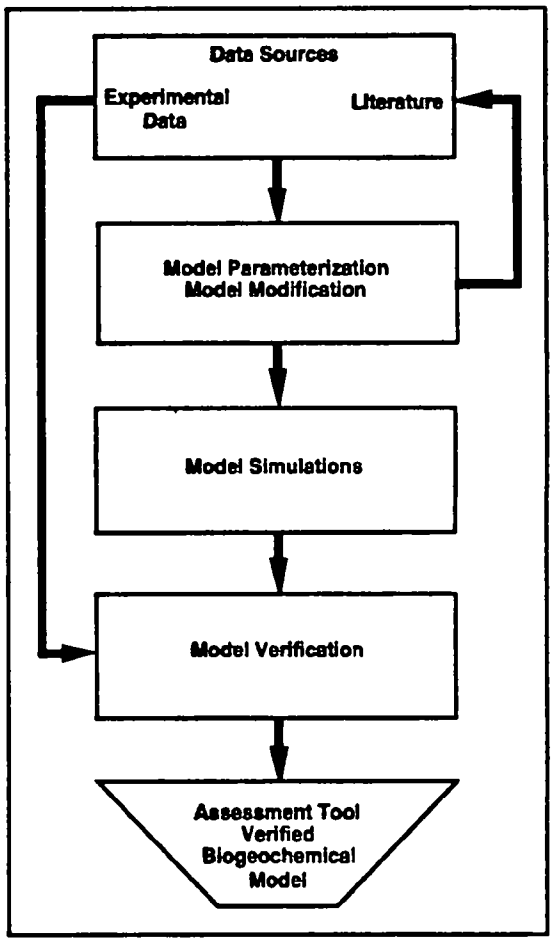

Figure 3-2. Illustration of the various steps and feedbacks that will be taken to develop a verified biogeochemical model for use as an assessment tool.

We have selected the Marine Biological Laboratory General Ecosystem Model (MBL-GEM) (Rastetter et al. 1991) as our primary working model because it most closely meets the preceding requirements,

i.e., it provides a process-based view of the acquisition, allocation, and chemical partitioning of C and N in plants and soils, and it can be calibrated using ecosystem- and regional-scale data. In addition, a preliminary parameterization of the model has already been established for Pacific Northwest forests (Appendix C).

Description of MBL-GEM

The MBL-GEM is a process-based model of C-N interactions in terrestrial ecosystems. Its structure (Fig. 3-3) is described in detail in Rastetter et al. (1991). The model is intended to be generally applicable to most terrestrial ecosystems and has been used in the past to analyze the responses of temperate deciduous forests, tropical evergreen forests, and arctic tundra to changes in CO_2 concentration, temperature, N inputs, irradiance, and soil moisture (Rastetter et al. 1991, 1992a, 1997; McKane et al. 1995, 1997a, 1997b).

The MBL-GEM simulates, at the stand level, photosynthesis and N uptake by plants, allocation of C and N to foliage, stems, and fine roots, respiration in these tissues, turnover of biomass through litterfall, and decomposition of litter and soil organic matter. The model currently simulates responses to changes in atmospheric CO_2, temperature, soil moisture, irradiance, and inorganic N inputs to the ecosystem. Carbon dioxide is lost from the ecosystem through plant and soil respiration. Inorganic N losses are assumed to be proportional to inorganic N concentrations in soil. The model calculates all changes on a monthly time-step. To be consistent with the time-step, monthly averages are used for all climate drivers.

Three major features of the model are important to our application. First, vegetation in the model acclimates to changes in the environment to maintain a nutritional balance between C and N (flexible within specified C:N ranges). Thus, environmental changes that stimulate photosynthesis (e.g., increased CO_2 or higher irradiance) result in an increase in allocation of C and N to fine roots, thereby stimulating N uptake. Similarly, environmental changes that stimulate N uptake (e.g., high inorganic soil N concentration) increase allocation of C and N to foliage, thereby stimulating C uptake. A second important feature of the model is that respiration rates of plant tissues are proportional to the amount of metabolically active N in those tissues (Ryan 1991). Thus, increased N availability increases productivity and growth in the vegetation, and it also increases the rate of plant respiration per unit biomass if the N concentrations in tissues increase. Finally, high N availability in soil stimulates the rates of decomposition for organic soil fractions like cellulose that have high C:N ratios. In effect, increases in N availability stimulate decomposition by facilitating the conversion of high C:N to low C:N products. This is consistent with evidence that increased N availability produces a "priming" or accelerating effect on overall rates of decomposition (Gill and Lavender 1983, Hunt et al. 1988).

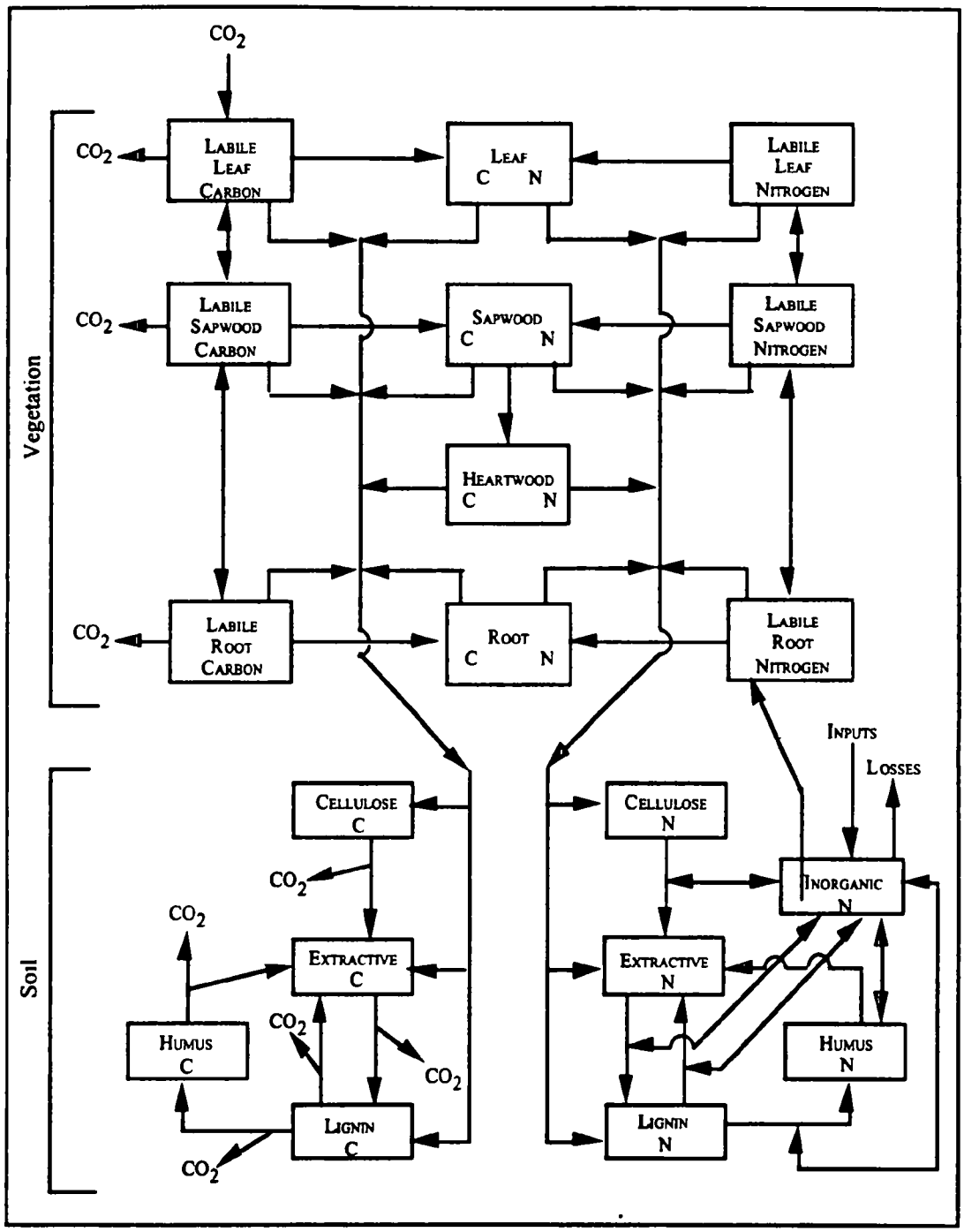

Figure 3-3. Schematic diagram of carbon and nitrogen cycles in the MBL-GEM (Rastetter et al. 1991)

Parameterization of MBL-GEM

The MBL-GEM is a lumped-parameter ecosystem model, meaning that it is a spatially averaged representation of ecosystem processes (e.g., Cosby et al. 1985). Because of the non-linearity of the process equations and because of the intense sampling required to characterize the spatial heterogeneity of most

ecosystems, lumped-parameter models are very difficult to parameterize from the bottom up (Beven 1989). That is, it is difficult to use fine-scale measurements of individual processes like leaf-level photosynthesis to parameterize the model (Rastetter et al. 1992b). Instead, the process equations in the model are "scaled" by calibrating them to data that are collected at the same scale at which the model is to be applied (e.g., ecosystem-level net primary production). The MBL-GEM does this by inverting the process equations so that the model parameters can be either directly calculated or estimated by least-squares from the system-level calibration data (this is equivalent to what Jarvis [1993] calls "optimization"). The rate constants for each C and N flux within the vegetation, the rate constants for humus turnover, and the C:N ratio of humus are calculated in this way.

This "top down" approach to parameterizing MBL-GEM is similar to the inverse modeling approaches used in geophysical and global C cycle modeling, where general circulation models and observed atmospheric CO_2 concentrations are used to estimate unmeasured CO_2 sources and sinks (e.g., Enting and Mansbridge 1989, Ciais et al. 1995). Although this approach is not often used in ecology it is extremely useful for our purposes, and for ecosystem science in general, because data for fine-scale processes (e.g., leaf-level photosynthesis) are often not available or are more difficult to obtain than data for coarse-scale processes (e.g., ecosystem-level net primary production). McKane et al. (1995 and 1997a) discuss in detail how this approach was used to parameterize the MBL-GEM for tropical evergreen forest and arctic tundra.

The data needed to parameterize MBL-GEM (Table 3-4) describe the distribution and fluxes of C and N among the various vegetation and soil compartments and concomitant changes in the environmental drivers. Because our goal is to derive a single parameterization of the model that is generally applicable to forests throughout the Pacific Northwest, we will obtain these data for a number of sites representing a range of age classes and climatic and edaphic conditions (Section 3.1 Site Characterization).

Methods used to collect data for parameterizing MBL-GEM

We will use the methods described below and in the various subtasks in Section 3.3 to obtain the vegetation and soil data from the intensive field sites needed for model parameterization (Table 3-4). Two categories of methods are described that represent intensive measures and simpler measures. The intensive measures are most accurate and will provide the primary data for model parameterization. The simpler measures will be developed in conjunction with the intensive measures, then applied to an extensive network of less intensively studied sites to evaluate their use as regional-scale indicators of ecosystem response to change. These indicators may be used for model parameterization when a measure is well-correlated to an intensive measure or, alternatively, they may be used for landscape-scale model validation when a measure provides a relative index of change among sites.

C and N Stocks

Vegetation C and N stocks.

Stem C and N. Stem C and N stocks (above and belowground woody tissues exclusive of fine roots < 2 mm in diameter) will be calculated by multiplying estimated stem biomass by measured stem C and N concentrations for each species. Total stem biomass will be estimated allometrically from measurements of tree diameter and height. The live (sapwood) and dead (heartwood) components of stem biomass will also be determined by analysis of tree-ring increment cores (intensive measure), or by the allometric relationship of sapwood area to leaf area index (simpler measure). The optimum number and size of sample quadrats at each field site will be established using methods described by Roberts et al. (1993). For our Cascade field sites, we estimated total stem biomass using a minimum of eleven 15 m x 15 m plots randomly located within 0.6 ha blocks.

Carbon and N concentrations of sapwood and heartwood, and all other plant and soil materials mentioned in this section, will be determined using gas chromatography methods (Carlo Erba) (USEPA SOP 3.01 *Carbon/Nitrogen Analysis*).

Leaf C and N. Leaf C and N stocks will be calculated by multiplying estimated leaf biomass by the C and N concentrations of live leaves collected at each site. Leaf biomass will be estimated from LAI as described under Subtask 1a. Estimated LAI will be converted to leaf biomass on a stand basis using the relationship between leaf biomass and specific leaf area for live leaf samples. Species' contributions will be weighted by their fraction of total stand basal area.

Fine root C and N. Fine root (<2 mm) C and N stocks will be calculated by multiplying estimated fine root biomass by measured C and N concentrations. Fine root biomass will be estimated using standard coring methods (Santantonio et al. 1977; see also Subtask 4a). At least ten 5-cm diameter x 20-cm deep cores will need to be collected per site (Vogt et al. 1981), and additional coring to 30 cm will be made for about half of these samples. Using these intensive measures as a reference, we will also investigate whether a simpler measure, or indicator, of fine root biomass can be developed using allometric relationships to leaf biomass and/or sapwood area.

Forest floor and soil C and N stocks

Coarse woody detritus C and N. Coarse woody detritus includes standing dead trees and fallen logs ≥ 10 cm diameter (Harmon and Sexton 1996) and will be sampled on the same 15 m x 15 m plots used for live stems (see above). All coarse woody detritus on each plot will be measured for length, large and small end diameters, and decomposition class (four classes ranging from no decomposition to highly decomposed). The C and N content of coarse woody detritus will be determined by calculating volume, converting volume to dry weight based on published wood densities by decomposition class (Grier and Logan 1977), and multiplying dry weight by measured C and N concentrations.

Table 3-4. Data needed to parameterize the MBL-GEM. Data marked with an asterisk (net N mineralization and soil respiration) are not essential for parameterizing the model but are important as a check on the calibration procedure (the model calculates rates for these processes to be consistent with mass balance requirements). The methods used to collect these data are classified into two categories representing intensive/difficult measures and simpler measures.

	Intensive/Difficult Measures	Simpler Measures (Indicators)
C & N Stocks (Mg ha^{-1})		
Vegetation C & N		
Stems (above and belowground wood)	Allometrically from diameter & height	Allometrically from diameter only
Sapwood/Heartwood	Tree increment core analysis	Allometrically from LAI
Leaves	Calculate from LAI & leaf mass/area	Allometrically from stem diameter
Fine roots (< 2 mm)	Soil core analysis	Allometrically from leaf biomass
Soil C & N		
Soil Organic Matter		
Litter layer (fine + coarse litter)	Destructive quadrat samples	Estimate from depth & bulk density
Mineral soil humus	Soil core analysis	
Inorganic soil N (ammonium + nitrate)	KCl extraction of soil cores	
C & N fractions (extractives, cellulose, lignin)		
Leaf, wood, and fine root litter	Chemical fractionation (subtask 5a)	
Soil humus	Chemical fractionation (subtask 7a)	
C & N Fluxes (Mg ha^{-1} yr^{-1})		
Net primary production C & N		
Stems (above- & belowground wood)	Tree-ring analysis/allometry	Dendrometer bands
Leaves	Litterfall traps	Needle age class analysis & allometry
Fine roots (< 2 mm)	(1) Root turnover, (2) N budget	
Litterfall C & N		
Stems (aboveground wood)	Annual total plot surveys	Annual line intercept surveys
Leaves	Litterfall traps	
Vegetation N uptake	Calculate from N requirement of NPP	
Leaf N retranslocation	Subtask 4b	
N leaching Lysimeters		

Net N mineralization*	Incubation of soil cores	Resin bags
Soil respiration*	Infrared gas analysis & Subtask 8a	
Other Data	**Intensive/Difficult Measures**	**Simpler Measures (Indicators)**
Leaf area index (projected m^2/m^2)	Subtask 4a	
Rooting depth	Visual inspection of soil cores & pits	
Environmental Data		
Atmospheric CO_2 concentration	Infrared gas analyzer	
Photosynthetically active radiation	Quantum sensors	
Air temperature	Temperature sensors	
Soil temperature	Buried temperature probes	
Soil moisture	Gravimetric & TDR methods	
Atmospheric N deposition	Dry & wet deposition collectors	

Forest floor C and N. The forest floor has been operationally defined for Pacific Northwest forests as the O1 and O2 horizons plus woody material < 10 cm in diameter (Harmon and Sexton, 1996). Because of the high spatial heterogeneity of forest floors in this region, about fifty 0.1 m^2 forest floor quadrats per site will need to be sampled at each site. Forest floor C and N content will be determined by multiplying ash-free dry mass per unit area by the concentrations of C and N. In addition to these intensive measures, we will investigate whether a simpler measure of forest floor biomass can be developed from depth and bulk density measurements.

Mineral soil C and N. Mineral soil samples will be collected at 20 cm increments to a depth of 1 m from several randomly located pits at each field site. Mineral soil C and N content will be determined using measured C and N concentrations, bulk densities, and gravel and stone contents.

C & N proximate fractions in litter and mineral soil humus (extractives, cellulose, lignin)

The chemical composition of soil organic matter is an important control on its decomposition and is represented in MBL-GEM as proximate fractions of extractives, cellulose, and lignin. The C and N content of each of these fractions will be determined for fresh plant litterfall (leaves and wood), fine roots, and soil humus using the procedure described by Ryan et al. (1990 (see Section 3.3, Subtasks 5a and 7a below).

C and N Fluxes

Net primary production.

Stem NPP. Stem NPP includes the annual increment of all above- and belowground woody biomass. Whereas tree-ring analysis can be used to estimate stem NPP for past years, dendrometers (expandable metal bands that track radial growth; Lassoie 1973; Keeland and Sharitz 1993; Telewski and Lynch 1991) are used to estimate current rates. For both methods, stem NPP is calculated using published allometric equations that relate stem biomass to diameter and height (Means et al. 1994). That is, stem NPP is the difference in total stem biomass before and after accounting for the measured diameter increment for 1 yr. We are estimating stem NPP for the past 10 years at our existing field sites by analyzing tree-ring samples for all trees over 1 cm dbh on the 15 x 15 m plots described above. Current monthly changes in stem NPP will be estimated using dendrometer bands attached at breast height (1.37 m). For these estimates, trees will be sampled by 20 cm diameter classes, with the number of samples per class being weighted according to the frequency distribution of tree diameters. Dendrometers provide a relatively simple and easily deployable method for measuring stem NPP across a number of field sites.

Leaf NPP. Leaf NPP for closed-canopy coniferous forests is approximately equal to annual dead plus live leaf litterfall. Because storm events may contribute to interannual variations in live leaf litterfall, leaf NPP estimates will be based on an average of 2-3 years of litterfall data. Live

and dead leaf litterfall will be determined from monthly samples of total litterfall at each site (see "Litterfall C & N" below). Using these intensive measures as a reference, we will also investigate whether a simpler measure of leaf NPP can be developed from the combination of leaf biomass estimates and leaf (needle) age class analysis.

Fine root NPP. We will estimate fine root NPP using two primary methods. One method is to divide measured fine root biomass (see "Fine root C and N," above) by the turnover time of fine roots measured in minirhizotrons. Minirhizotrons have already been installed at the low and high Cascade field sites, and new fine root production and mortality (Tingey et al., 1996, 1997) have been measured monthly since the spring of 1995 and will continue (Section 3.3, Subtask 6a). We will also estimate fine root NPP using the N budget method of Nadelhoffer et al. (1985). That is, by subtracting the N requirement of leaf and stem NPP from annual net N mineralization (see below), the remaining amount of net N mineralization can be used to calculate fine root NPP. Using both of these methods should provide a means of cross checking our data.

Litterfall C and N

Litterfall includes all aboveground fine (leaves, cones, woody material < 10 cm in diameter) and coarse (woody material $\geq$ 10 cm in diameter) detritus production. Fine litterfall will be measured using fifty randomly located 0.3 m^2 screen-lined traps at each of the forested sites (Grier and Logan 1977). We will measure coarse litterfall at our intensively studied field sites by annually resampling the 15 m x 15 m coarse woody detritus plots (see above). At less intensively studied sites, coarse litterfall will be measured using the line intercept surveys annually (Harmon and Sexton 1996), a less accurate but much faster method. The C and N content of all fine and coarse litterfall components will be determined by multiplying dry mass by measured C and N concentration.

Vegetation N Uptake

Vegetation N uptake will be calculated as the N requirement of above- and belowground NPP. The basic calculation is [NPP x N concentration for leaves, wood, and fine roots] minus leaf N retranslocation.

N leaching losses

Leaching loses of ammonium-N, nitrate-N, and dissolved organic nitrogen (DON) will be measured using porous cup lysimeters buried at a depth of 1 m. Soil solutions will be sampled at monthly intervals or as soil moisture conditions dictate. The concentrations of ammonium-N, nitrate-N in solution will be analyzed colorimetrically using autoanalyzer methods (Technicon 1977, USEPA 1979). DON will be determined as the difference between total dissolved nitrogen (TDN) and inorganic N, with TDN being determined by persulfate oxidation followed by colorimetric analysis for nitrate-N (D'Elia et al. 1977, Currie et al. 1996). A hydrologic model parameterized for the Cascade forests (Marks and Dozier 1992) and precipitation data collected at the intensive sites will be used to estimate total leachate volume so that total N losses can be estimated.

Net N mineralization

Net N mineralization will be measured using *in situ* incubation of intact soil samples within 20 cm deep PVC tubes covered with thin polyethylene film (Hart et al. 1994). The length of the incubation period may be as short as a month or as long as over winter depending upon objectives and changing environmental conditions. Soil samples taken before and after incubation are extracted with 1 M KCl and colorimetrically analyzed for nitrate and ammonium (Technicon 1977, US EPA 1979) (SOP # 3.10 *Alpkem Autoanalyzer*). Net N mineralization is estimated as the difference between the amount of total inorganic-N (NO_3^- + NH_4^+) accumulated within the core after the incubation period relative to the amount present in the soil prior to incubation.

Ion-exchange resin bags (IER bags) buried a few centimeters below the mineral soil surface have been shown to be suited for comparative studies of N availability among sites or treatments (Binkley and Matson 1983, Binkley 1984, Binkley et al. 1986, Giblin et al. 1994). The accumulation of NH_4^+ and NO_3^- on the anion-cation exchange resin is regulated by the processes of mineralization, immobilization, plant uptake, and transport; however the method cannot distinguish the relative importance of these processes. And, unlike the closed-tube method, resin bags do not provide an areal estimate of net N mineralization because an unknown volume of soil is sampled. The IER bag method has shown to produce results that correlate highly with other methods for measuring N-mineralization, are less spatially variable than other methods, and correlate with NPP and fine root biomass (Binkley and Matson 1983, Binkley et al. 1986). We believe that the strengths of the IER bag method are particularly suited for the development of an indicator for assessing N availability or mineralization rates across sites and years (e.g., Binkley and Hart 1989, Giblin et al. 1994).

We will prepare the resin bags and extract the ions according to the methods described in Binkley et al. (1986). IER resin bags will be buried at about 5 cm beneath the top of the mineral soil in each study site adjacent to the locations used for measuring N mineralization with soil cores. Bags will be left in the field for up to a year (actual length of time will be determined through a series of preliminary experiments). The ions will be extracted with 1M KCl, and the ammonium and nitrate analyzed as above.

Soil Respiration

Soil respiration is the total flux of CO_2 from the soil surface to the atmosphere and includes both root respiration and microbial respiration. Soil respiration rates can be most accurately measured by infrared gas analysis (Nay et al. 1994). We are measuring soil respiration rates at our intensive field sites by placing a portable infrared gas analyzer (IRGA) over PVC collars permanently installed in the soil (Soil respiration SOP# 7.10 *In situ Soil Respiration (Li-Cor 6200): Field and TERA*). The IRGA-PVC collar system gives reliable measures but is labor intensive, especially for developing diurnal and seasonal respiration patterns. An automated system of measuring soil respiration is also being developed (Section

3.3, Subtask 8a), which will provide data for cross-checking the results obtained here.

Other data

Rooting depth. In addition to estimating fine root biomass (see "Fine root C and N" above), the maximum depth of fine roots will be determined by examining deep soil cores or pits excavated at each site. Maximum rooting depth will be operationally defined as the point above which 95% of visible fine roots occur.

Environmental Data

The environmental drivers for MBL-GEM include atmospheric CO_2 concentration, photosynthetically active radiation (PAR), air and soil temperatures, soil moisture, and N deposition. These data will be obtained from several sources, including data from the climate sensors installed at our intensively studied sites (Section 3.1, Table 3-2), regional and site-specific data from state (e.g., Oregon Climate Service, University of Oregon) and U.S. Forest Service, historical temperature and precipitation data reconstructed from tree-ring analyses, and future climate scenarios predicted by general circulation models (e.g., the Goddard Institute of Space Studies GCM). Data for wet deposition of N will be obtained from the National Atmospheric Deposition Program (NADP) site at the H.J. Andrews LTER established in 1979. Few data are available for dry deposition of N, however, a summary of the data available for the western U.S. indicates that dry deposition can be approximated using a 1:1 relationship for dry to wet deposition (Young et al. 1988). Additional dry deposition data will be obtained from USEPA CASTNet sites (US EPA 1995) and the California Air Resources Board (Blanchard et al., 1996).

Model evaluation and verification

We will use the parameterized MBL-GEM as an assessment and predictive tool to determine the effects of environmental change on ecosystem C and N dynamics, to identify additional potential sensitive indicators of C and N dynamics, and to link changes in the potential indicators identified in Section 3.4 (below) to likely stressors. For example, how will predicted increases in CO_2, temperature, and N deposition during the next century affect net primary production, soil respiration, and N transformations and loss from forest ecosystems? Or, how will periodic harest of forestsaffect the long-term productivity and sustainability of forest ecosystems (see appendix C)? In these applications the model will be used to predict long-term changes that are outside the range of any possible validation data set except, of course, by waiting the requisite time for the full ecosystem response to develop. Obviously, waiting is ill-advised when one must assess potentially significant changes in the environment. Therefore, confidence in these models must be built through the accumulation of relatively weak corroborating evidence and tests. Of the four categories of evidence or tests that Rastetter (1996) proposed for evaluating models of ecosystem response to environmental change (see above), only three tests are useful for our purposes and are described below. Although none of the tests can be used as severe and crucial tests of long-term (decades to centuries) model predictions (Rastetter 1996), a conflict

with any of these tests will lead to a re-examination of the model parameterization or the model itself.

Tests using short-term experimental data:

Model predictions of short-term (seasonal to less than a decade) ecosystem C and N dynamics will be evaluated using the data collected in this research plan that describes seasonal and interannual variations in photosynthesis, NPP, litterfall, soil respiration, soil N mineralization, and N leaching. While these data cannot address slow-responding processes and feedbacks that influence long-term ecosystem response, they are extremely valuable for understanding the transient responses of ecosystems to acute disturbances such as harvesting, insect outbreaks, fire, N deposition, etc. These transient responses may be of primary interest in some cases, for example, in predicting nutrient losses to "downstream" ecosystems following forest harvest.

Space-for-time substitutions:

Model predictions of long-term ecosystem C and N dynamics can be evaluated using two types of space-for-time substitutions. The first type of space-for-time substitution uses chronosequences of plots to examine successional changes in ecosystem characteristics. In this way, space (location of the plot) is substituted for time (time since disturbance). We will examine chronosequences of forest stands in the Pacific Northwest following two major types of disturbance, fire and harvest, to test how well the model simulates post-disturbance recovery of vegetation and soils.

A second type of space-for-time substitution can be used to evaluate long-term predictions of the effects of climate on ecosystems. In this case, if the climate at one location is expected to change so that it resembles the climate at another location, then the ecosystem characteristics (e.g., C and N stocks) at the original location might be expected to change so that they resemble the present-day characteristics of the second ecosystem (Rastetter 1996). This type of space-for-time substitution has been used to test the equilibrium predictions of MBL-GEM for projected increases in temperature across the Amazon basin (McKane et al. 1995). Similarly, we will test the predicted long-term (equilibrium) adjustment of Pacific Northwest forests to projected changes in temperature and precipitation using data describing C and N cycling in old-growth forests at a number of sites throughout the Pacific Northwest, for example, sites at the H.J. Andrews Experimental Forest (Grier and Logan 1977, Sollins et al. 1980) and the Middle Santiam Wilderness (Fujimori et al. 1976).

Comparison with other models:

Although comparisons among different models cannot substitute for tests against real-world data, they can be used to evaluate the relative importance of various processes in determining long-term responses to environmental change. Agreement among models means that the results of one model do not conflict with the principles underlying the other models. The degree of confidence obtained from such agreement depends on how well the underlying principles in each model have been established and how independent

these principles are among the models (Rastetter 1996). For example (Rastetter 1996), one model might be based on biogeochemical principles of mass balance and the interactions among C and N cycles (e.g., MBL-GEM) and another might be based on the principles of competition among individual trees for light and soil resources during succession (e.g., forest succession models; Section 3.3.2). If the two models agree on the accumulation rate of carbon in the vegetation, then the biogeochemical model has not conflicted with the constraints of the succession model, and the succession model has not conflicted with the constraints of mass balance and C-N interaction in the biogeochemical model. Alternatively, when the model results are not in agreement, important controls on ecosystem response not included in one or all of the models may be identified (Ryan et al. 1996). We will compare the predictions of MBL-GEM against those for a forest succession model (Section 3.3.2), as well as other biogeochemical models (e g., CENTURY). We anticipate that these comparisons will be extremely useful in guiding further development of the MBL-GEM.

Task 2: Modeling at the landscape scale

The main goals of this task are to: (1) link the MBL-GEM and a coupled energy and water balance model (Daley et al. 1994, Marks and Dozier 1992) to common spatial data on land cover/land use, terrain structure, and soil characteristics to determine the effects of differences in landscape pattern on model outputs at watershed scales, (2) use the product of goal 1 to predict the effects of anthropogenic stressors on forest landscapes of different pattern, and (3) develop relationships between forest landscape spatial patterns and ecological processes. This research task will examine those ecological processes whose changes are most likely to be influenced by changes in spatial and temporal patterns of forested ecosystem landscapes (i.e., changes in land use/land cover) and by other anthropogenic stressors such as change in climate and air pollutants. Initially these ecological processes will include water quality, net primary production, and C and N balances.

The distribution and pattern of natural and human dominated systems in landscapes influence ecological processes which in turn affect production of goods and services that landscapes provide humans. Changes in pattern of landscapes can significantly affect their response to other anthropogenic stressors such as air pollution and global change, thus affecting the input and output of materials (e.g., water quantity and quality [O'Neill et al 1977], soil and sediments), regional productivity, or biodiversity. Certain configurations of ecosystems are likely to make some landscapes more vulnerable to stressors than others. Although the linkages between landscape structure and function have been subjects of interest in ecological research for some time (Bormann and Likens 1979), recent advances in the developing field of landscape ecology has provided a basis for studying relationships among landscape structure, pattern, function, and change (Forman and Godron 1986; O'Neill et al. 1988; Turner 1989). Advances in remote sensing and subsequent data analysis in a GIS have facilitated analyses of major ecological

processes in relation to landscape pattern and composition. Indicators reflecting changes in land cover and/or land use are likely to be useful in monitoring large-scale changes in the function of forested ecosystems if relations can be documented.

Methods

Linking the MBL-GEM to landscape-scale spatial data bases

Initially, we plan to use the South Santiam drainage (10 sub-watersheds of different land use/land cover patterns and climatic gradients) as a test area for this objective, followed by a scaling up to the Western Cascades. As described above (section 3.1.2), digital data for land cover/land use and terrain structure are available for the entire South Santiam system.

The energy and water balance model (Marks and Dozier 1992) provides daily outputs of precipitation, snowmelt, evaporation, surface and soil temperature, incident and net radiation, runoff, soil moisture, and available moisture. These outputs and the land cover/land use data for the South Santiam sub-watersheds will serve as input drivers to the MBL-GEM. Soil C and N content and texture are also important input drivers of the MBL-GEM. We plan to use the Willamette National Forest Soil Resouce Inventory as the starting point for this work, and collect additional soil samples for quantification of soil C, N, and texture as needed. A parameterized MBL-GEM will then be simulated, pixel by pixel, for each land-cover / land-use class in the sub-watersheds using the appropriate climatic and soil moisture regimes generated by the energy and water balance model and soil C and N status from the soil surveys for the same pixels. The MBL-GEM-simulated outputs of NPP, N losses, and C balances will be summed by sub-watershed. These simulated outputs will be used to investigate relationships between landscape pattern and ecological processes as described next.

Predicting the effects of anthropogenic stressors on forest landscapes of different pattern

This objective will build on the previous one, in that the landscape-scale MBL-GEM will be simulated under a range of possible scenarios of climate change, change in atmospheric composition, and increased N deposition. Changes in the outputs of NPP, N losses, and C balances for each subwatershed under these different scenarios will be compared to indices of landscape pattern (described below) to investigate how the pattern influences the vulnerability of landscapes to further change.

Development of relationships between forest landscape spatial patterns and ecological processes

The ability to quantify landscape pattern and structure is requisite to understanding landscape function and change over time. For this purpose we will use a versatile public domain computer program, FRAGSTATS, which produces a comprehensive array of landscape pattern metrics (McGarigal and Marks 1995). The software is automated for use with either vector or raster images on either Unix or PC platforms. We will examine a variety of possible indicators of landscape spatial pattern of different land cover/land use classes, including various area measures,

patch density, size and variability, as well as edge, shape, core area, diversity, contagion, nearest neighbor and interspersion metrics. We will also examine percent distribution of forests in the various seral stages as a measure of landscape vegetation structure.

We will use the sub-watersheds of the South Santiam watershed and their 1991 land cover/land use data (Section 3.1.2). The map data will be analyzed with FRAGSTATS to quantify landscape pattern in the sub-watersheds, detecting differences based on land-use, land cover, and forest management practices. The metrics of landscape pattern will then be related to MBL-GEM outputs described above to determine watershed-level functional responses to climatic and edaphic factors, forest management practices, and other stressors.

To understand how spatial patterns in the South Santiam subwatersheds influence water quality measures (nutrients, other chemical ions, and microbiological contaminants), we will institute a regular water sampling program for the subwatersheds near their mouths over a period of at least 3 yr. Using field sampling methods adapted from the EMAP Surface Waters Program (US EPA 1997b), we will collect grab samples monthly and analyze for nitrate, ammonium, dissolved organic nitrogen, dissolved organic carbon, sulfate, chloride, iron, calcium, magnesium, potassium, sodium, turbidity, conductivity, pH, alkalinity, and presence of various microorganisms. To calculate mass movement of the various chemical species, stream flow will be measured. Monthly, seasonal, base-flow, and peak-flow bulk movement of nutrients and other elements in stream water will be calculated as the product of water concentrations and discharge. The water quality values will be statistically compared with landscape pattern metrics as detailed above.

Water quality impacts of forest management activity have been demonstrated under drastically different conditions, such as comparing paired watersheds where one is intact old-growth forest and another is entirely clear-cut (e.g., Sollins et al. 1981). Much less has been reported on the impacts on water quality when more subtle differences in forest management are compared. In order to understand what differences in management practices and landscape patterns are necessary to cause significant changes in water quality parameters, we will investigate nested South Santiam sub-watersheds at different spatial scales. Five sub-watersheds of near-equal size (~ 5 000 ha) will be sampled. In addition, paired sub-watersheds of approximately 500, 50 and 5 ha will be monitored. The smaller sub-watersheds will provide greater opportunities for observing greater differences in forest management, landscape pattern and seral stage distribution.

Monitoring will be accomplished by obtaining continuous records of stream discharge and automating collection of as much water quality data as feasible. Weirs may be installed with depth sensors to aid in estimating instantaneous discharge. Electronic sensors and data loggers will be installed for monitoring parameters such as water temperature, conductivity, turbidity, dissolved oxygen, and pH. Anions, cations,

dissolved inorganic and organic nitrogen, dissolved organic carbon, ANC, total phosphorus, and several microbiological measures will be determined for grab samples collected less frequently (e.g., bi-weekly).

Combining the water quality data with metrics of landscape pattern and seral stage distribution will enable us to answer the question of how much difference is necessary to detect changes in water quality. Conversely, we will be able to estimate the extent of management disturbance required to produce adverse impacts on forest streams.

3.2.2. Forest succession modeling

Task 3: Forest succession model for stand to landscape

Objectives and background

The objective of this task is to enhance the applicability of forest succession models in montane areas generally, and in the Pacific Northwest (PNW) specifically for use in forest indicator research and assessments. The goal is to develop, test and use models reliable enough to be integral components of forest indicators of ecosystem vulnerability to environmental change. From the suite of potential field data collection and model building activities which can enhance forest succession model validity (Bugmann and Solomon, 1997), we chose as the first task, the creation of a model of tree mortality, both as a forest indicator in its own right, and for incorporation into a montane forest succession model. This task requires documenting specific processes important in the mortality of trees. Our second task is to increase the accuracy of the method used to estimate environmental limits of trees species, which underlies the basic climate response of the forest succession models. This task requires documenting fine-scale distributions of climate variables and tree species geography.

Forest structure obviously will respond to directional shifts in environmental variables. Natality and mortality rates of individual tree species will change. Gaps in forest canopies, and loss of whole forests, are likely to result because death of tree populations requires only a few years while establishment and regrowth of replacement trees requires several decades to centuries (Solomon, 1986; Kirschbaum and Fischlin 1996). This lag in replacement of dying trees should be statistically detectable at regional scales. However, it will be more difficult to produce definitive measures of related changes that are specific to any given locality, such as the Cascades study sites of this proposal. The competitive relationships among adjacent trees in a stand will be modified as e.g. warming increases growth of some and decreases it or leaves it unaffected in others. Eventually, certain species will be outcompeted entirely and will disappear from individual forest stands, local watersheds, then regions. The trees with the poorest success under chronic climate stress are likely to be late-successional species, that is, those with the longest life cycles, with the slowest growth rates, and with the greatest shade tolerance (Solomon and Leemans 1990, Solomon et al. 1993), a set of properties which also characterize tree species capable of the slowest responses to climate change.

In addition to different tree species, different stages of tree life cycles also are differentially vulnerable to environmental variations. The middle-aged trees present in a forest canopy are most resilient (Lorimer and Frelich 1984, Peet and Christiansen 1987). Those approaching senescence are much more vulnerable, and newly-germinated seedlings are the most vulnerable of all (Harcombe 1987). Hence, the establishment phase of late-successional species should provide the most sensitive indicators of forest structure response to chronic environmental change. Yet, the establishment of tree seedlings is also the most variable. Western hemlock, a late-successional tree species characteristic of the Pacific Northwest, may produce 2 X 107 seeds per ha each year which may reduce to 2 X 104 saplings 20 years later (Packee 1990). However, fewer than 2 X 102 trees reach maturity, even in the absence of stand-replacement disturbances. Moreover, establishment "pulses" at tree range boundaries may be decades to centuries apart (Savage et al. 1996, Arseneault and Payette 1997). Documenting the sensitivity of seedling establishment to changing climate would require many thousands of samples over many years in many locations for each tree species. Therefore, this project will focus upon the age-dependent mortality of late-successional trees as the most appropriate indicator of directional changes in forest structure.

Although tree mortality is less vulnerable than seedling establishment to environmental variations, it has certain advantages as an indicator of long-term directional changes in forest structure and function. Mortality is an unambiguous state which varies in response to stress and age (Waring, 1987). With seeding establishment, it defines long term forest dynamics (Harcombe, 1987), making inclusion of mortality considerations critical to the application of most forest indicators. Mortality rates under differing past environmental conditions can be objectively defined (e.g., Henry and Swan, 1974) and departures from them by other mortality events and trends can be measured and assigned statistical probabilities of occurrence. Large-scale mortality (die-back) is an obvious condition which possesses considerable political importance, and which sometimes can be measured with inexpensive remote sensing techniques, permitting rapid assessment of the statistical characteristics needed to attribute cause to the mortality rate changes. When included in forest succession models, mechanistic mortality models can permit calculation of forest community-level tree deaths related to environmental change.

Strong advantages are also found in forest succession models themselves for forest indicator research. Particularly pertinent is their ability to capture and predict the year-to-year changes to be expected in forest structural characteristics (e.g., changing size and age distributions of individuals from each species), and to do so as a function of yearly variations in temperature, soil moisture, CO_2, ozone, and other (changing) environmental properties. Their weaknesses for our purposes includes their non-mechanistic treatment of stress-induced mortality, the inaccurate species-climate correlations permitted by available environmental data in montane areas, and their inability to

predict the dynamics on any specific plot or set of plots without very detailed information on plot history (e.g., Solomon 1988). These weaknesses will be treated in the research proposed below.

Nature of forest succession models

Forest succession models (also referred to as gap or stand models) mimic the dynamics of tree establishment, growth and mortality by multiple species of differing ages in a gap created by the death of a dominant tree in an otherwise continuous forest canopy (Fig. 3-4). There, the models simulate interspecific competition for sunlight, water and nutrients based on individual species differences in shade tolerance, drought tolerance, and nutrient requirements. They simulate the vertical characteristics of tree density on a (usually) circular plot of specified size and they calculate the amount of light which reaches each vertical level as a function of the leaf areas above that level (Fig. 3-4, center). They assume that the maximum dimensions of each tree species (maximum diameter, height and age), previously measured in the field, are also the maximum dimensions each species could reach under ideal environmental conditions (light, warmth, soil moisture, and nutrients). Each simulated year, climate conditions (Fig. 3-4, right margin) are calculated as random variables from documented means and standard deviations, and reduce growth of trees on the plot from the growth maxima which rarely if ever occur either in nature or in the models.

Both tree establishment and mortality of established trees, of interest here, are treated as stochastic processes (Fig. 3-4, left margin). Mortality, for example, is modeled as a constant probability of death such that only 2% of established trees reach their maximum known age. A second stochastic process provides that diameter growth below a threshold at any age produces an enhanced mortality probability (usually a 1 in 3 chance of mortality during the subsequent 10 years), which is most likely in the youngest (i.e., smallest diameter increment) trees and in the oldest (i.e., slowest growing) trees. These stochastic rules may simulate much more rapid elimination of trees under changing climate, than rates of tree loss under the actual mechanisms which control tree mortality. This problem becomes a critical model flaw if tree mortality rates are to be reliable indicators of changing forest condition.

Climate constraints on mortality, as well as on growth and reproduction, are defined by the coincidence of mapped boundaries of tree species with mapped climate variables. From these overlays, one defines such extreme climate parameters as the minimum temperature of the coldest month or the maximum annual days of soil moisture below wilting point, found within the geographic range of each species. Recent reviews of the capability of forest succession models to represent probable forest responses to future climate change (e.g., Bonan and Sirois 1992, Loehle and LeBlanc 1996, Schenk 1996) condemn this underlying methodology. The critics suggest that accidents of Holocene tree migration have produced a current geographic range of species ("realized niche," more correctly "realized range") which is smaller than the geographic range the species potentially could occupy

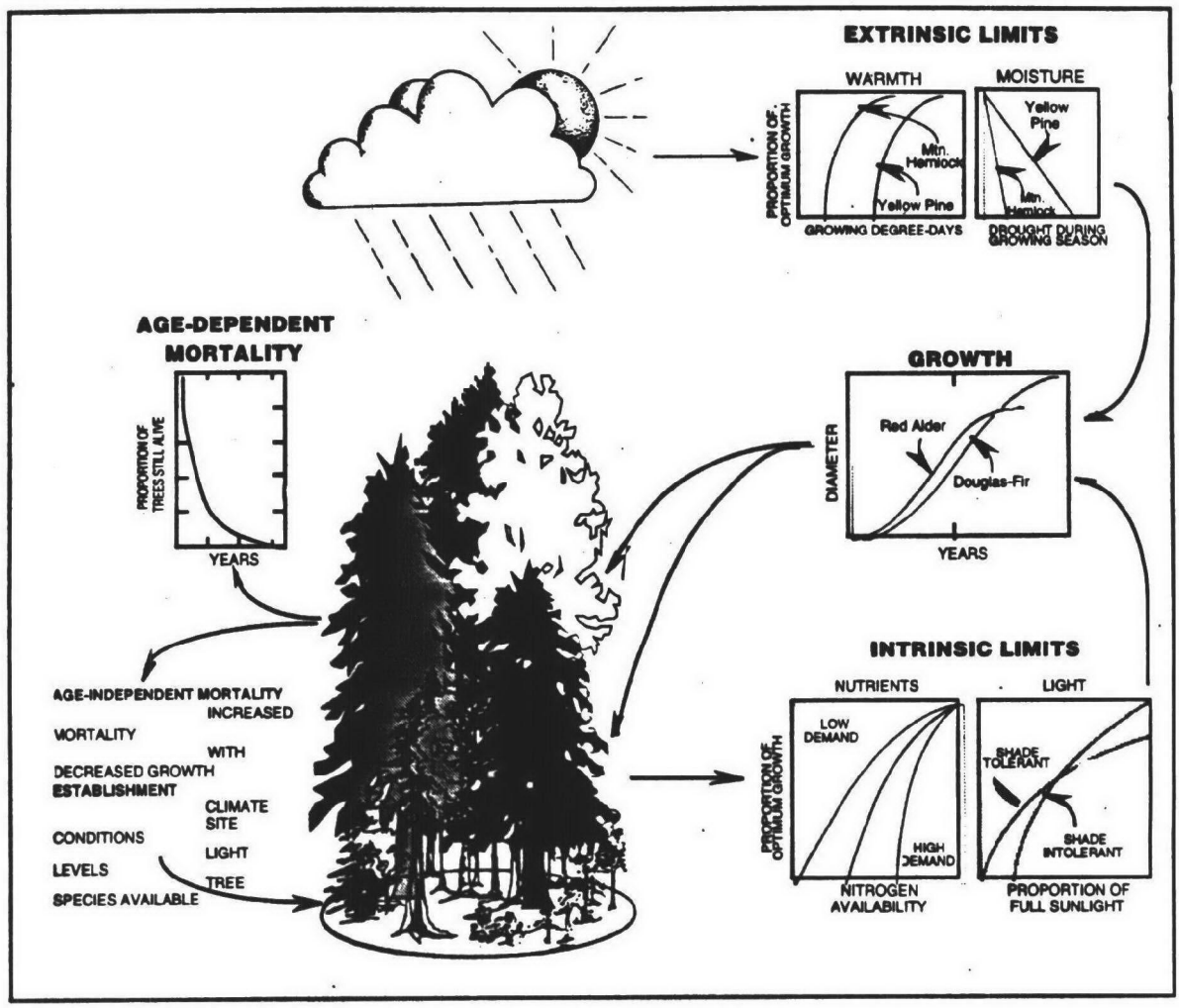

Figure 3-4. Conceptual diagram of the forest succession model ForClim 2.9, after adaptation of a generic temperate-zone model (Bugmann and Solomon 1995) for forests in the Pacific Northwest (Bugmann and Solomon 1997).

("fundamental niche" or range), introducing a false climate-growth correlation into succession models.

This criticism may (or may not) be appropriate in flat or gently rolling terrain. However, Tsukada (1982a, 1982b) has showed that species ranges in mountainous terrain have not been subject to delayed or obstructed migration paths because of their close (up or downslope) proximity to suitable habitat under even rapid past climate changes. Hence, the montane forests of interest in the PNW should be much less prone to the realized versus fundamental range problem.

However, implementation of the climate-species correlations in the PNW encounters more direct methodological restrictions. Growth of montane tree species at their boundaries, where extreme climate parameters are documented, is restricted to a few suitable habitats (e.g., cold north or warm south-facing slopes, cool, damp valley bottoms), rather than to average

conditions which are described by regional climate measures. Similarly, climatic gradients are so steep in mountainous terrain that the entire range of climate values found within a tree species' range can be telescoped within the few kilometers between adjacent climate stations at different elevations. This scale problem is the specific focus of proposed research described below.

FORCLIM forest succession model

Gap models have been applied in Pacific Northwest forests for at least 15 years (Hemstrom and Adams 1982, Dale and Hemstrom 1984, Kercher and Axelrod 1984, Urban et al. 1993, Burton and Cumming 1995). Each model version varies in capability and validity, none being well-suited to the problem of regional indicators, even in the absence of the weaknesses discussed above. Variables and parameters in the ZELIG model (Urban et al. 1993) have been calibrated to work for the H. J. Andrews LTER Site (ZELIG2.PNW; Stephen Garman, pers. comm. 1997), but documentation has not been published to date, and the value of one site to represent the PNW is not great.

The ForClim model (Bugmann 1994) simulated forests of Europe and eastern North America (Bugmann and Solomon 1995) with equal accuracy. It was modified (V2.9) to incorporate the peculiarities of forests of the Pacific Northwest (Bugmann and Solomon 1997). ForClim 2.9 has been tested for its ability to reproduce stand biomass and species composition on a transect of 27 sites from the Oregon Coast, eastward across the Coast Range, the Willamette Valley, the Cascades, and into the cold desert near Bend, Oregon. The model also has been tested against tree size distributions and species composition at three elevations (500 m, 1000 m, 1400 m) in the H. J. Andrews LTER Site. These tests reveal that ForClim reproduces PNW forest dynamics more accurately than other available forest succession models, and that the ability of the model to reproduce eastern North American and European forest composition and biomass also is improved by the PNW modifications.

The new modeling developments on which we can focus our necessarily limited resources include the current the lack of mechanistic stress-related mortality processes, and the inability of the model to relate species geography to the very steep temperature and moisture gradients found in the western mountains. The former problem (mortality processes) will require field data and literature synthesis on natural mortality rates. The latter problem (species geography) will require additional data on geographical distributions and associated environmental conditions under which individual tree species are found. Some of these data are available by careful examination of the published literature, and some will require field excursions to record previously undocumented occurences. The methods used to reduce both of these weaknesses will coincidentally permit application of the models to specific locations, by generating the historical data (tree-ring data) on mortality with which to replace the succession model's stochastic estimator of these data.

Methods

The data we will collect to improve the FORCLIM model to the point that it more accurately reproduces the spatial distributions and temporal sequences of species composition, density and size/age distributions in the Pacific Northwest are in two groups: one group is required to develop the mechanistic mortality model and a second group is required to improve the way in which species' environmental limits are derived. The following information describes the research activities we propose to implement to reach these specific goals.

Data for constructing a mechanistic mortality simulator

In addition to use of the scientific literature to create a mortality simulator, we will collect data sets on mortality in the PNW to parameterize and test the simulator. Three methods will be applied. One approach is to examine stand remeasurement data from permanent plots, where available. These data frequently include measurements of the mortality since the previous survey, and may extend over several decades, from samples collected at regular or irregular intervals. Harcombe (1986) utilized such records from seven remeasurements during 50 years at 130-year old forests from Cascade Head Experimental Forest in the Coast Range near Otis, Oregon to define temporal sequences of mortality, and their sources, during stand development there. We will determine the availability of appropriate remeasurement plot data in the Pacific Northwest forests of Oregon, Washington and Idaho, and will define mortality rates either by our analysis of these data, or by encouraging mortality analyses by those with responsibility for the data

A second means to examine the rates and changes in mortality is by reconstruction of forest site histories (e.g., Henry and Swan 1974, Franklin et al. 1981). The approach is based on the slow rates of deterioration of fallen dead trees in the Pacific Northwest (Harmon, et al. 1986) combined with the ability to crossdate the outer growth rings of long-dead trees with the inner rings of recently dead or living trees (e.g., Smiley and Stokes 1968, Cook and Kariukstis 1990). The primary method consists of developing crossdated dendrochronologies, mapped for all the dead standing and downed trees in several stands or study areas, and the calculation of the mean and variance of mortality rates for the areas represented by the several data sets (Franklin et al. 1981). We propose to reconstruct mortality histories of several stands in the Toad Creek Road and Falls Creek intensive study sites, where some tree ring chronologies have already been collected and processed. In addition to our documenting of long-term mortality rates for development of a mortality simulator, we will use the resulting measurements to provide important background mortality and tree growth information for parameterizing stand mortality in the forest succession simulation verification exercises discussed above. The mortality measurements and site histories will also support the other above- and below-ground research at the intensively-studied sites. Because of the labor-intensive nature of these field data collections, we expect to generate data from only three or four 0.2 ha plots at each of the two sites.

A third, very different means to calculate mortality rates, is to measure the change in growth rates recorded by the last decades of annual growth rings in dead trees which occur in larger diebacks. This approach establishes species-specific growth variations which may predict imminent mortality (Schwiengruber et al. 1986, LeBlanc et al. 1987), and which can then be used to hypothesize specific mortality cause and effect. These regional dendrochronologies provide a spatial contrast to the site-specific forest site histories described immediately above. We propose to implement this approach by developing dendrochronologies (2 cores from each of 20-40 trees per site) at 10-20 sites in Cascades forest stands which have died back (e.g., the Blue Mountain dieback; the Santiam Pass dieback), and in adjacent still-living stands of the same species, in order to document the growth patterns in the decade or so immediately preceding the diebacks (e.g., Swetnam 1987). Although this research activity is considerably more exploratory than other work proposed here, we believe its potential for defining productive mortality indicators is quite high.

Data for enhancing accuracy of species-environment relationships.

A fundamental problem to implementing gap models in the Pacific Northwest involves the difficulty of defining environmental tolerances of species from their co-occurring spatial distributions in montane regions of very steep environmental gradients. Distributions both of tree species and of climate variables are mapped too coarsely to permit accurate estimation of the tolerance values. We propose to resolve the problem of steep elevational gradients through use of fine-resolution digital elevation models (DEMs). The DEMs will be used to disaggregate the range of topographic values represented in spatial units of the finest-resolution climate data base available. These data will define a fine-scale set of climate data required to drive the stand models (monthly growing degree days, temperature of coldest month, tri-monthly soil moisture and snow status), and will be matched with field observations of species distributions in the areas of their geographic ranges where they reach critical limits.

Because local temperature and soil moisture depend on elevation, exposure, and topographic position, these physical variables will be critical targets to calculate and map from a DEM (500 m resolution) within each grid square (VEMAP data containing 4 km x 4 km pixels) represented in the climate data base (Dodson and Marks 1997). Distributions of topographic variables will be transformed to proxy climate variables using topography-climate relationships (e.g., solar insolation and sun angle for temperature versus topographic position; known seasonal lapse rates and humidity for temperature versus elevation; PRISM model for moisture versus elevation [Daly et al. 1994]).

In addition to the fine-scale mapping of proxy climate variables, we will also document the relationships between climate and actual local tree distributions (approximately 50 species are currently found in the Oregon Cascades or are appropriate for survival there under global climate change scenarios). Review of the literature, visits with local forest experts,

as well as field inspections will be conducted in regions where tree species reach a critical geographic limit. We envision several two-week excursions in which we visit multiple locales representing east-west and north-south range limits of the most common of the 50 species. The objective will be to determine the frequency and fidelity with which species are segregated in specific topographic situations (e.g., on north-facing slopes and in deep valleys at the southernmost edge of their range; on south-facing slopes and on calcareous soils at the north edge of their range; etc.) in these non-optimal areas.

The proxy climate data and the local distributional data will be used to define the actual range of environmental conditions in which each species can survive within its current geographic range. The proximity of widely differing conditions in montane areas should have permitted most available niches to fill during the past several thousand years since inception of the current climate (Tsukada 1982a, 1982b). Hence, the realized range (current range limits) of these montane species is probably little different from the fundamental (potential) range beyond which climate actually limits species' presence.

The relationship between climate and distributions of 50 tree species, generated by the foregoing, will be independently tested in part by examining relationships between tree ring indices and climate at 20 to 100 sites scattered throughout the geographic ranges of two of the most important tree species of the Pacific Northwest: coastal Douglas fir (*Pseudotsuga menziesii* var *menziesii)*, and Pacific western yellow pine (*Pinus ponderosa* var. *ponderosa*). Douglas fir occupies cool, mesic sites along the Pacific coast some 2200 km from central British Columbia to central California. The Pacific variety of western yellow pine occupies warm, dry sites from southern British Columbia almost to Mexico. The elevational ranges of the two species at any given location overlap, with dominance determined where fire favors ponderosa pine and fire suppression favors Douglas fir. The analysis will require us to collect tree ring chronologies from the entire latitudinal N-S range of the two species, as well as from selected low to high elevation transects, each chronology containing paired sites consisting of climatically stressed and non-stressed trees.

We will identify old-growth stands of trees, and adjacent weather stations with long station histories, the set of which represent the range of conditions throughout the geographic ranges of Douglas fir and yellow pine. Tree ring indices will be developed, based both on tree ring widths and on late-wood density, and will be used to create response surfaces to characterize the response of tree rings to individual climate variables as they vary with elevation and latitude (e.g., Cook and Cole 1991, Thompson et al. 1998). The response surfaces in turn will provide a measure of tree growth response to climate independent of the DEM surface analyses. This will be particularly valuable as a means to testing our assumption of equivalence between the realized and fundamental range of these two species.

The tree-ring analyses proposed here have not been used for this purpose in the past, and hence, may not provide the model verification required for indicator-level reliability. However, if we can demonstrate the technique to be effective, we expect to collect and analyze data on additional tree species important in the Pacific Northwest stand models. In any case, the development of a permanent tree-ring analysis capability at WED will coincidentally lead to development of stand histories at the Cascades research sites. Such histories will be. a critical component of our analyses comparability of research results from these sites.

3.3. Resource utilization

The previous section (3.2) described modeling approaches that will be evaluated and/or developed for assessing impacts and predicting changes in forested ecosystems from natural and anthropogenic stressors and management practices. This section will focus on measurements of C and N utilization by trees (3.3.1) and metabolism of C and N in the forest litter and soil (3.3.2). The Resource Utilization Tasks will serve several functions including:

- Providing data for model parameterization, calibration, evaluation, and modification
- Providing fundamental data on key ecosystem processes and their response to various stressors
- Evaluating the suitability of various ecosystem processes as indicators to assess the current condition of forested ecosystems
- Developing indicators of ecophysiological processes and complexity and function of the soil foodweb to assess the current status of forested ecosystems.

These tasks will focus on determining the natural spatial and temporal variability of key ecophysiological processes and the complexity and function of the soil foodweb (Fig. 3-5) at intensive and extensive field sites. Field sites will be selected that are appropriate for testing specific hypotheses regarding resource utilization (see section 3.1.2). For example, retranslocation of N and nitrogen use efficiency as a function of site nutrient status is being investigated in tree species. To accomplish this, three sites are being used that have differing nitrogen availability. Two of the sites differ fourfold in N, and are at the same elevation and approximately the same climate. A third site has higher N availability but is at a lower elevation and a differing growing season length. Specific hypotheses and the use of field sites with appropriate climate, nutrient, etc gradients will enable interpretation of variance in the measures of carbon and nitrogen allocation.

Many of the measurements described in this section will also be made in the experimental facilities at WED where we will measure the same ecological processes and components under known stressors (see Section 4).

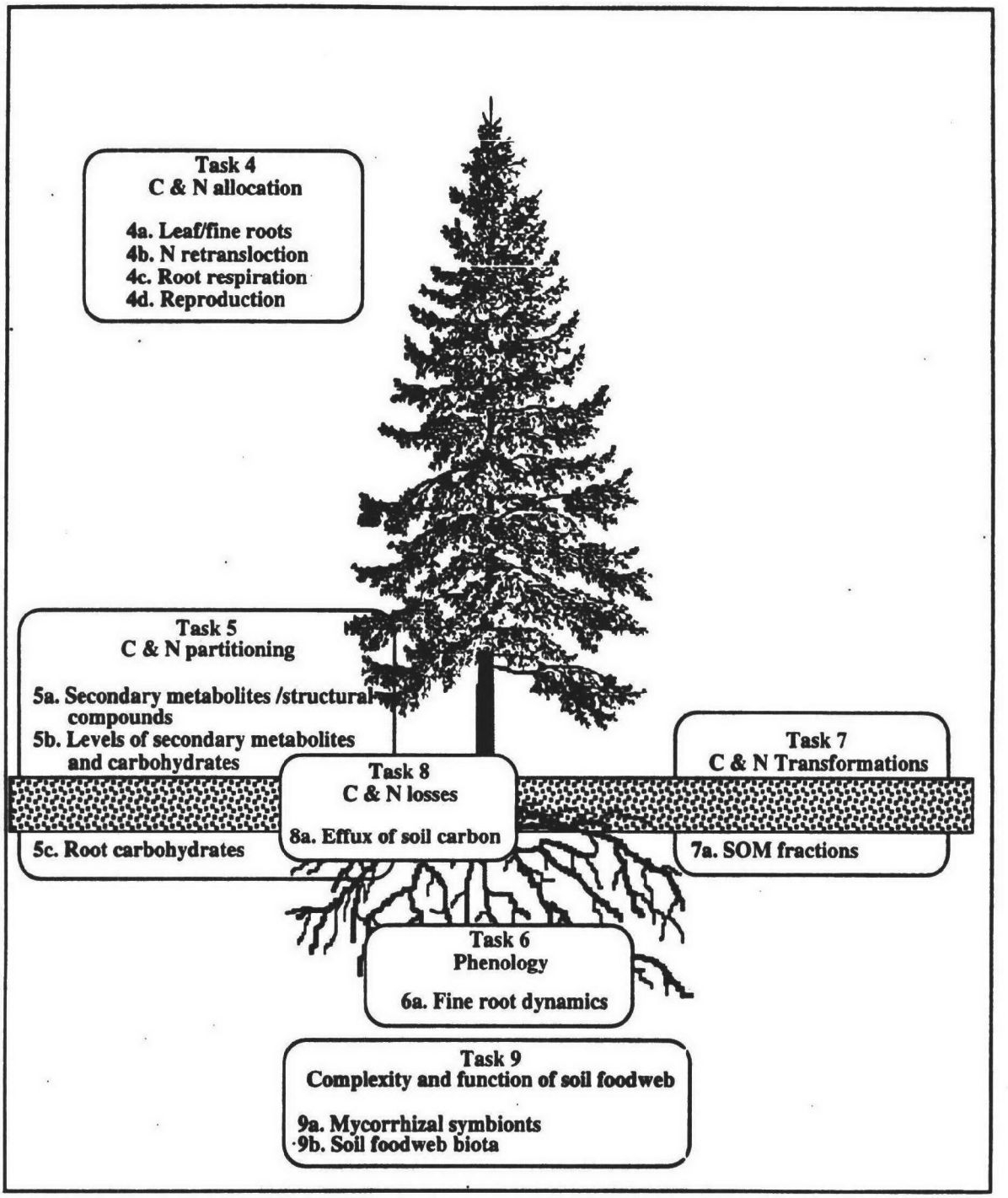

Figure 3-5. Identification of the research tasks to measure spatial and temporal variability of resource utilization (C and N) by producers and consumers.

3.3.1 Resource utilization-producers

Task 4: Carbon and Nitrogen Allocation

Subtask 4a. Leaf area/fine root biomass ratios as measure of C allocation

Objectives and background

The main goal of this task is to provide fundamental data on C and N allocation between leaves and fine roots, including determining the interannual variation in the LAI/fine root biomass ratio. The ratio will be compared to climate records and data on soil moisture, temperature, and C and N content to determine the sensitivity of the ratio to various environmental and edaphic factors. This task will also contribute several key types of data needed for the biogeochemical model (Section 3.2.1). Light extinction data will be used to estimate LAI for the stands and to provide information on light extinction important for calibrating MBL-GEM. The fine root biomass data will contribute to estimating the C and N pools of the fine roots and also will be used to determine the rooting depth of the stand.

Leaves and fine roots play significant roles in resource acquisition. Consequently, the allocation of C and N to these organs to maintain a balance between C and water and nutrient acquisition is important for a plant to grow, reproduce, and to permit it to adapt to changing environmental and edaphic conditions. For example, C fixation occurs in leaves but its rate depends on their N content and thus on N uptake via root activity. Because the chemical composition of a plant can vary only within narrow limits, a proper balance between the functioning of leaves and fine roots must occur. The functional balance concept (Brouwer 1962, 1983) implies that fine roots and leaves continually adjust their relative resource allocation patterns as resources change. For example, Lou et al. (1994) proposed that the fine root fraction would be stable when the N supply matched the photosynthetic supply but if the CO_2 stimulation of photosynthesis was larger than shoot growth (limited by nutrient shortage) the excess photosynthate would stimulate root growth.

To study the allocation of C and N, we will follow the recommendation of Körner (1994) and use a three compartment (needles, woody tissue and fine roots) model to describe carbon allocation and functional relationships in plants. The three compartment model is the same as that used in the MBL-GEM model (Rastetter et al. 1991) which is being used in the biogeochemical cycling modeling portion of the research (3.2.1).

The ratio between leaves and fine roots (biomass or area) has been used to investigate the impacts of various treatments on C allocation. In ponderosa pine, N fertilization initially increased the LAI/fine root biomass ratio. However, this effect decreased as the plants grew, possibly because the relative abundance of N to plant size decreased (Tingey et al. 1996). Pregitzer et al. (1995) also found N fertilization increased the LAI/fine root ratio in poplar. Both Norby et al. (1992) and Körner (1994) reported that elevated CO_2 increased fine root mass but had no

effect on leaf mass leading to a decrease in leaf mass/fine root mass ratio. However, in ponderosa pine, the LAI/fine root length ratio was unchanged by elevated CO_2 indicating that the relationship between nutrient absorbing surfaces and photosynthetic surfaces was not changed by CO_2 exposure (Tingey et al., 1996); Pregitzer et al. (1995) found a similar trend for poplar.

Methods

Leaf Area Index (LAI) and leaf biomass

LAI will be estimated from measures of light extinction made below the canopy (Pierce and Running 1988, Gholz et al. 1991). The light extinction method is: (1) site specific and (2) provides information on light extinction important for calibrating MBL-GEM.

A Licor light bar (1 m) will be used to measure light extinction at permanently marked locations along a transect in the forest at each site. Measurements in the clearcuts will serve as the reference for top-of-canopy light levels. All light measurements will be made within one hour of solar noon near the time of the summer solstice to minimize the effect of shadows from tree stems. Beer's Law will be used to estimate leaf area from the light extinction data (Jarvis and Leverenz 1983). Leaf area will be then converted to leaf biomass on a stand basis using measures of the specific leaf area (g dry weight m^{-2} leaf area). The light extinction method is preferred over allometric methods of estimating leaf biomass because it is site specific.

Fine root biomass

Fine root biomass will be estimated using standard coring methods (Santantonio et al 1977). At least 20 locations will be sampled per site using cores 5 cm in diameter and 30 to 100 cm deep. Soil and roots will be separated using wet sieving methods. Roots will be separated into coarse (>2 mm) and fine root (≤2 mm) fractions, dried and weighed. Core samples will be collected at 3 mo intervals to determine seasonal changes in fine root biomass.

C and N allocation between leaf and fine root pools

Measurement of leaf tissue C and N pools is described in section 3.2.1 (*Leaf C and N*). Oven-dried and weighed (corrected to an ash-free basis) roots will be ground to pass a 40 mesh screen, and analyzed for C and N concentration using a flash-combustion gas chromatography method (Carlo Erba Standard Operating Procedure (SOP) 3.01 *Carbon/Nitrogen Elemental Analysis*). Carbon and N pools will be calculated by multiplying fine root biomass by measured C and N concentrations.

Subtask 4b: Nitrogen and phosphorus retranslocation and resorption efficiency/proficiency

Objectives and background

The objectives of this subtask are to determine (1) the potential resorption of N and P (maximal withdrawal of each nutrient from senescing foliage), the resorption efficiency (percent reduction of nutrient between green leaves and senesced leaves), and the resorption proficiency (measured level of nutrient in senesced foliage) in 1-2

dominant tree species over a period of 3 yr and across a characterized gradient of growing conditions; and (2) values needed to parameterize MBL-GEM (see section 3.2.1).

Nutrient resorption, and in particular N resorption from senescing leaves before abscission takes place, is one of the most important processes for plants to conserve nutrients. Because senesced, falling leaves account for >70% of litter, the efficient retranslocation of nutrients from leaves into storage tissues in stem or root is an essential process in both the individual and the ecosystem. A number of factors can cause realized resorption of nutrients to be less than potential resorption in some years, including water availability (Escuedero et al. 1992), timing of abscission (Killingbeck et al. 1990) and shade (Chapin and Moilanen 1991). Anthropogenic stresses, such as tropospheric ozone, could also affect the resorption process. For example, ozone causes premature leaf fall in several trees species (e.g., Keller 1988, Wiltshire et al. 1993, Pell et al. 1995, Weber et al. 1997). If ozone-induced abscission proceeds before the resorption process is complete, this could disrupt nutrient conservation in individuals and possibly cause changes in nutrient availability from litter.

Millard (1996) summarized several nitrogen budget studies in evergreen and deciduous trees showing that internal cycling of N (i.e., N conservation) is a major source of N providing up to 90% of this nutrient used in seasonal growth. In addition, as trees get larger their N uptake rate decreases but their N storage capacity and reliance on internal nitrogen cycling increases (Millard 1996). Disruption of the N resorption process reduces plant fitness in subsequent years. May and Killingbeck (1992) demonstrated substantial reductions in foliar biomass (41%), radial stem growth (54%) and fruit production (90%) in a 3 year study of oak when nutrient resorption was blocked. Late season drought causes early leaf abscission in deciduous trees and disruption of normal nutrient resorption, and is a contributing factor to reduced stem growth in mature trees (Killingbeck et al. 1990) and a selective pressure in changing community species composition.

Killingbeck (1996) distinguished the measured differences in resorption efficiencies (percent reduction of a nutrient between green and senesced leaves) as differences in resorption potential of the species or temporal differences in realized resorption. The measurement of the potential resorption (the maximum withdrawal of nutrients from foliage) is not possible directly; however, estimates of the degree to which realized resorption approaches potential resorption in individual species can be achieved by developing a knowledge of the levels to which the species can reduce nutrients in senescing leaves (resorption proficiency) (Killingbeck 1996). This task proposes to take the multifaceted approach suggested by Killingbeck (1996) to develop a measure evaluating N cycling in different site conditions and over time as a potential indicator of tree (individual to population) condition or as an input variable into an indicator of forested ecosystem condition.

Methods

Measurements of concentrations of N, P and Ca in presenescence and senesced needles of ponderosa pine, Douglas-fir and western hemlock will be made on composited needle samples at selected dates after determining the time period when concentrations of these nutrients are relatively stable (maximum and constant) for each species. Samples of presenescence senesced needles will be collected from a range of diameter classes for each of the three species.

Needle area and dry weights will be measured to calculate nutrient content. Content is calculated as the product of specific leaf mass (g cm^{-2} of needle) and N or P concentration (µg gm^{-1} or ng gm^{-1}). Total N will be measured on dried and ground needle samples using the Carlo Erba C and N analyzer (SOP 3.01). Total P and Ca will be measured on dried, ground and digested tissue using an ICP spectrophotometer (William 1984). The content of Ca is determined for comparison because Ca is not resorbed.

Resorption efficiency will be calculated for each tree in each sampling period as the difference in N or P content (µg cm^{-2} of needle) between presenescent and senesced needles, divided by presenescent needle N or P content. Temporal changes in N, P, and Ca concentration will be determined and related to changes in estimated needle mass.

Subtask 4c: Root respiration

Objectives and background

The goal of this subtask is to examine (1) the use of stable C and O_2 isotope ratios in quantifying specific root respiration, as well as area-specific microbial activity, and (2) the utility of such a measure as an indicator of C dynamics in evaluation of forest condition.

Previous studies have found that soil CO_2 flux increases from soils containing plants exposed to ozone stress, and the increase occurs before any changes in growth are observed, suggesting that a change in soil C flux may be an early indicator of stress (Andersen and Scagel 1997; Scagel and Andersen 1997). One critical component of soil CO_2 flux is root respiration. Root respiration is important both for understanding plant response to stress and for characterizing system level C fluxes. Because C flux from soil occurs both from plant roots and decomposition of soil organic matter, it is important to discriminate between the two. Isotopic C and O_2 ratios offer a means to discriminate between the two C flux sources because each has a unique isotopic signature (Lin et al. 1998). We can quantify the source of the CO_2 released from a known volume of soil by knowing the isotopic characteristics of the C and O_2 sources. This approach allows us to quantify specific root respiration (per gram root), as well as area specific microbial activity. The subtask will compliment the isotopic work being conducted to deconvolute soil-system CO_2 fluxes (see subtask 8a).

Two approaches are envisioned that will use stable carbon isotope ratios. The first will involve characterizing the δC13 ratio in root total nonstructural carbohydrates (TNC) and in root structural components. By knowing isotopic ratios of C in TNC and structural root components, we will be able to better understand the conditions under which the carbon was fixed and later transported to the root. While not a stress-specific indicator, it does provide an integrated measure of conditions present during carbon acquisition. The second approach will be to measure the various components of soil respiration. Knowing the C and O_2 isotopic signature of each soil component will allow us to calculate the contribution of each component of the soil flux to the total δC13 signature of the CO_2 collected from the soil surface, including fine root respiration (this will be accomplished under subtask 8a).

Methods

Characterizing the δC13 ratio in root TNC:

During the first year, large lateral roots will be identified and traced to donor tree (initially Douglas firs of several size classes). Soil will be removed from around a portion of the root, and samples will be taken. Roots in three different diameter classes will be sampled: > 2 mm, 5-20 mm, and > 10 cm. Large diameter roots (>10 cm) will be cored with a 1 cm diameter increment corer rather than by excavation. Samples will be frozen on dry ice for transport to the laboratory, and subsequently stored frozen until they are lyophilized (Andersen et al. 1991). Freeze-dried samples will then be ground, and analyzed for TNC (Wilson et al. 1995) and carbon isotopic signatures.

Characterizing the δC13 ratio in soil plugs:

A trench will be dug approximately 3-5 m from the base of each of 6 mature trees, in an area where root proliferation is evident. To obtain a baseline value, soil-surface CO_2 flux will be measured in undisturbed soil adjacent to the trench using a Licor 6200 (Andersen et al. 1997a) and using gas collection apparatus. A horizontal plate will then be driven into the soil, parallel to the soil surface, at a depth of approximately 30 cm. Using the same location as baseline values, a PVC pipe (15 cm diameter) will be driven into the soil surface directly above the plate until it reaches the plate at a depth of approximately 30 cm. This process will result in isolating an intact cylinder of soil. Soil surface CO_2 will again be measured using a Licor and the gas collection device. After CO_2 collection, the intact plug will be removed from the soil and taken to the lab for analysis. The soil plug will be separated into litter, roots, soil organic matter, and mineral soil, and dried to quantify the mass of each component. Dried soil components will be analyzed to determine their carbon isotopic signature. Knowing the carbon isotopic signature of each soil component will allow us to calculate the contribution of each component of the soil to the total δC13 signature of the CO_2 collected from the soil surface. Using mass balance equations (Lin et al. 1998), the contribution of each component of the soil flux to the total respiratory flux will be obtainable, including fine root respiration. By comparing values before and after cylinder

insertion, it will be possible to evaluate whether significant changes in CO_2 flux resulted from wound response of severed roots in the soil column.

Subtask 4d: Reproduction

Objectives and background

The main goals of this sub-task are to (1) Quantify seed viability, as a measure of reproductive potential, for dominant tree species, and (2) quantify stoichiometric relationships of C and N allocation to wood, needles, and seeds. Quality of reproductive tissues plays a significant role in forest resource allocation and the perpetuation of tree stands and communities from one generation to the next The life of a seed is a complex series of events from the initiation of fruiting through the dispersal of the mature seed and germination (Krugman et al. 1974). Fruit, cone, and seed production and viability are affected by various combinations of physiological factors, weather, insects, diseases, predation by birds and mammals, and anthropogenic stressors (Burns and Honkala 1990; Harper 1977). We believe that an understanding of seed quality, and relative allocation of C and N to tree wood, needles, and seeds (Waring and Schlesinger 1985) has potential as a sensitive indicator of forest condition.

Seed viability

Tree climbers will collect samples of ripe cones, as well as associated branches and needles, from the three species in August and September each year at the Falls Creek and Toad Creek Road sites. Twelve Douglas fir cones per tree will be collected from different heights and sides of ten randomly selected mature (of seed-bearing age) trees at each field site. Branch and needle tissue will also be collected at two of the 12 cone collection locations on each sampled tree. Sixteen western hemlock cones per tree will be collected from four mature trees at each site. Branch and needle tissue will also be collected at two of the 16 cone collection locations on each sampled tree. Thirty-two Pacific silver fir cones per tree will be collected from eight mature trees at the Toad Creek Road site only. Branch and needle tissue will also be collected at two of the 32 cone collection locations on each sampled tree. Seeds will be tested by a recognized tree seed research laboratory (i e., USDA Forest Service's Dorena Tree Improvement Center) by both X-ray contrast (Moore 1969) and germination procedures for reproductive capacity.

Carbon and nitrogen allocation

C and N concentrations will be determined on oven-dried subsamples of the seeds, branches, and needles collected as described above. The tissues will be ground to pass a 40-mesh screen and analyzed for C and N using a flash-combustion gas chromatography method (Carlo Erba SOP 3.0.1 *Carbon/Nitrogen Elemental Analysis*).

- ***Task 5: Carbon and Nitrogen Partitioning***

Subtask 5a: C partitioning to secondary metabolites and structural compounds

Objectives and background

The main goals of this sub-task are to (1) quantify the partitioning of C into secondary metabolites and structural compounds in leaf and fine root tissues of the dominant trees to determine how the quantities vary by forest successional state, climatic factors, and by season and (2) use the results to contribute to the parameterization and verification of the MBL-GEM (Section 3.2.1).

The C fixed by plants is generally allocated for growth, reproduction, storage, and defense (Waring and Schlesinger 1985, Tuomi et al. 1988). Major determinants of plant tissue quality are an array of chemical and structural compounds, i.e., secondary metabolites, lignin, and cellulose (Coley et al. 1985, Lindroth et al. 1993) that serve as defenses against herbivory and pathogenic infection (Coley et al. 1985, Waring and Schlesinger 1985). Secondary metabolites and structural compounds in both above and below ground tissues also influence the quality of the organic matter inputs to the soils, and subsequently its decomposition (Swift et al. 1979, Horner et al. 1988) and quantity of soil organic matter. The nature and quantity of secondary metabolites in plant tissues are determined by the availability and utilization of resources in the local environment (Coley et al. 1985, Lindroth et al. 1993), which in turn are affected by other stressors such as air pollutants and global change (Lawler et al. 1997).

Research shows that polyphenols (e.g., tannins, terpenes) and fiber in leaves significantly increases with increasing leaf lifetimes on the tree. The types of available resources in an environment also place constraints on the types of secondary metabolites plants make. For example, in N-limited environments one would expect higher concentrations of C-based chemicals such as tannins or phenols than N-based chemicals such as alkaloids (Coley et al. 1985, Lindroth et al. 1993). Recent studies have shown that conditions in the environment that increase the C:N ratio in leaves, such as low soil N or increased atmospheric CO_2, significantly increased the amount of C partitioned to secondary metabolites (Lindroth et al. 1993, Lawler et al. 1997). However, to what extent environmental stressors that reduce C fixation affect the concentration of secondary metabolites and structural compounds is unknown. Further, increased atmospheric N deposition has the potential to alter C:N ratios of leaf tissues (Asner et al. 1997) and thus affect the quantity and nature of the secondary metabolites.

We propose that the concentrations of secondary metabolites and structural compounds have the potential to serve as indicators of forest condition. Any change in the quantity and composition of leaf tissue is likely to change a forest's susceptibility to herbivory and pathogenic infection. Moreover, changes in the quantity and composition of secondary metabolites and structural compounds of both leaf and fine root tissues are likely to affect the structure and function of the soil food web, which in turn will likely affect organic matter decomposition and nutrient mineralization.

Methods

Samples of leaf and fine root tissue will be collected from the dominant trees in the intensive study sites. Tissue samples will be collected four times a year — winter, spring, summer, and fall months — for at least 3 yr. Once a seasonal pattern has been established in the intensive sites, tissue samples will also be collected from the extensive sites in subsequent years for further evaluation of this potential indicator. The timing of leaf and root sampling will be coordinated with other sampling schemes to maximize tissue collection efficiency. Leaf samples will be collected from about ten different diameter trees at each site, and separated into new and old needles. Samples of fine roots will be collected in coordination with subtask 1a (above).

All tissue samples will be dried at 50 C and ground at 0.85 mm mesh size (Ryan et al. 1990). We will use the methodology recommended by Ryan et al. (1990) for determining the concentrations of secondary metabolites, lignin, and cellulose. This methodology is a sequential treatment of the sample using the wood products techniques for polar (simple sugars, and polyphenols) and non-polar (fats, oils, and waxes) extractives, which we assume are surrogates for secondary metabolites, and the forage fiber techniques for cellulose and lignin (Ryan et al. 1990). The polar extractives will be further analyzed for simple sugars by a colorimetric method (DuBois et al. 1956) and for phenols by the Folin-Denis colorimetric method (Allen et al. 1974).

Subtask 5b: Levels of secondary metabolites and carbohydrates

Objectives and background

The main goals of this subtask are to determine (1) the amount of within plant, within community and between community variability in nonstructural carbohydrates and secondary metabolites and (2) if the level of nonstructural carbohydrates and secondary metabolites varies in response to stress.

Secondary metabolites are formed from photosynthate that is not used for construction or maintenance, and one primary function appears to be protection against predation by animals and microorganisms (Waring and Schlesinger 1985). The synthesis of secondary metabolites may be costly to the plant, requiring a steady flow of precursors from primary metabolism, enzymes and energy rich co-factors (ATP, NADPH, etc.) (Harborne 1993) unless they are being used for storage. This cost has led to at least three different but not mutually exclusive theories on their evolution (Tuomi 1992).

1) The optimal defense theory assumes that there is a limited amount of resources that a plant can devote to defense and that there are alternative demands for these limited resources (Rhoades, 1979). The theory concerns evolutionary trade offs between growth and defense (Tuomi 1992). The theory predicts that 1) commitment to defense should decrease in the absence of enemies and increase when plants are subjected to a high risk of attack, 2) plants should evolve defenses in inverse proportion to the cost of defense and 3)

plant parts most valuable in terms of fitness should be most effectively defended. Therefore; environmentally stressed plants should be less well defended against pathogens and herbivores than unstressed plants.

2) The carbon-nutrient balance (CBN) theory suggests that carbon-based metabolites will be positively correlated with the carbon-nutrient balance and conversely that nitrogen-based metabolites will be negatively correlated with this balance (Bryant et al. 1983). Bryant et al. (1983) suggested that phenotypic responses of secondary metabolism are governed by a carbon-nutrient balance. Moderate nutrient stress should in fact enhance carbon-based defenses. CBN theory emphasizes site differences in resource availability as a factor in differences in secondary metabolite within a species. Therefore, moderate nutrient stress should enhance carbon-based defenses.

3) The growth-differentiation balance (GDB) theory suggests the existence of a physiological trade-off between growth and differentiation with the latter term including secondary metabolism synthesis (Tuomi et al. 1990). The theory has perennial plants divided into two groups; 1) growth dominated plants with rapid growth, poor chemical defense but with a highly inducible resistance system, and 2) differentiation-dominated plants with a slow growth rate, well defended with high levels of toxin but with poorly developed inducible resistance (Harborne 1993). GDB theory emphasizes temporal variation in resource availability. Rapidly growing plants should be characterized by lower and more plastic defense levels than slow-growing plants. The theory is based on three assumptions; 1) that genetic correlations between growth and defense are negative, 2) that growth is more sensitive than photosynthesis than to specific environmental stresses, and 3) that growth dominated plants have more plastic defenses than differentiation dominated plants (Tuomi 1992).

There is limited experimental evidence in support of these theories. However, Waring and Pitman(1985) were able to demonstrate that lodgepole pines susceptibility to mountain pine beetle decreased with the lessening of intraspecific competition or the increasing the availability of nitrogen. This work supports the optimal defense theory. It suggests that the lowest priority was the allocation of photosynthate to protective chemicals and therefore a plants first response to environmental or abiotic stress should be a shift of carbon allocation away from secondary metabolism. Other work by Reichardt et al. (1991) tends to support the CBN model when using carbon based secondary metabolites with low potential turnover rates. The lack of consensus and experimental evidence on the response of secondary metabolites to stress suggests there is a need for further investigation. .

In addition, plants may respond to different stresses including ozone (Matyssek er al. 1992), herbivory (Ayers 1984) and disease (Farrar 1992) by the accumulation of nonstructural carbohydrate in their leaves or stems. Therefore, it is plausible that high levels of leaf nonstructural carbohydrate and low levels of constitutive protective chemicals could

be an early indicator or predictor of an ecosystem in decline. In some cases, especially were the majority of the protective chemicals contain nitrogen, changes in the C/N ratio in leaf tissue may be a simple but sensitive indicator.

Methods

In order to answer the first goal of quantifying the amount of variability at several scales on the levels of nonstructural carbohydrates and secondary metabolites in plants, several field sites along the Sanatiam corridor will be sampled. Suggested sites would include the current Falls Creek, Soap Grass, and Toad Creek sites. Plant species common to the sites will be used to determine nonstructural carbohydrates and secondary metabolites in the roots and leaves. Plant species of interest are Douglas-fir (Pseudotsuga menziesii), western hemlock (Tsuga heterophylla), Oregon grape (berberis nervosa), salal (Gaultheria shallon) and bracken fern (Pteridium aquilinum). Douglas-fir and western hemlock were chosen because they are common dominant forest trees in the Pacific Northwest. The other three species are common understory or pioneer species following forest disturbance. The species are taxonomically diverse and have different secondary metabolic compounds. The results will determine the between site variability of similar communities composed of the same species.

Sampling of tissues will coincide with changes in plant phenology. This will determine the amount of seasonal variability within and between species and sites. Trees will be sampled prior to bud break, during the expansion of new growth, fully expanded growth and after bud set. Needle samples will be taken by one year age classes up to three years at three height levels in the canopy and in all four cardinal directions. Herbaceous plants will be sampled as new growth emerges, when fully expanded and prior to tissue death in the fall. On plants with over wintering leaves tissue samples will be taken prior to bud break, also. Root samples will be harvested from each plant at the same time as foliar samples are taken. Initially five plants of each species per site will be sampled in order to determine within community variability.

All tissue samples will be analyzed for non-structural carbohydrates using High Performance Anion-Exchange Chromatography. Samples will be analysized for total carbon and total nitrogen using flash combustion methodology. Trees will be analyzed for condensed tannins (Hagerman 1987; Julkunnen-Tiitto 1985), bracken fern for cyanogens (Jones 1988), and salal and Oregon grape for alkaloids (Keeler 1975)

Once the amount of within plant, within community and between community variability has been characterized, the same plant species will be used to address the second goal of determining what effects stress has on the level of nonstructural carbohydrates and secondary metabolites in plants. Differences in response to nitrogen levels will be determined between the Toad Creek (low soil nitrogen) and Soap Creek (high soil nitrogen) sites. Differences in light quantity between shaded and unshaded bracken plants in the clear cuts at the Falls Creek and Toad

Creek sites. Eighty percent shade cloth will cover some of the fern plants while others will remain in full sun. The bracken ferns are commonly connected by rhizomes. Areas around each fern sample area will be trenched below the rhizome level and back filled after sheet metal or plastic sheeting has been place in the trench for a root barrier. Some plants of salal and Oregon grape will be protected from herbivory and pathogens by spraying with pesticides using a back pack sprayer or fenced from large herbivores. Salal and Oregon grape plants not sprayed with the pesticide will be sprayed with an equal volume of water.

The amount and timing of tissue sampling will be determined from the results of the first goal. The types of analysis will be the same as described under the first goal.

Subtask 5c: Root total nonstructural carbohydrates (TNC)

Objectives and background

The goals of this subtask are to determine (1) the seasonal patterns of TNC accumulation and use; 2) the optimum time to measure concentrations; and 3) the appropriate root diameter class to measure. Root carbohydrates may be useful as an indicator of current and past tree "status" because TNC represents an important reservoir of energy for the tree during times of stress (Waring and Schlesinger 1985). Trees store carbohydrates throughout the plant to maintain respiratory pools and to provide energy for new growth in the spring. Any stress that reduces the plants ability to store TNC may reduce the plants ability to deal with stressors such as insects or fungal pathogens, poor nutrition, or anthropogenic in origin (Johnson 1989, Tingey and Andersen 1991).

To establish root TNC as a useful indicator, historical and seasonal trends need to be well documented for each forest species of interest. In addition, tree age affects TNC status and, therefore, sampling across a range of age classes will be required to provide information on the spatial and temporal variability of this potential indicator.

Methods

During the first year, large lateral roots will be identified and traced to donor trees. Soil will be removed from around a portion of the root, and samples will be taken. Roots in three different diameter classes will be sampled: <2 mm, 5-20 mm, and > 10 cm. Large diameter roots (>10 cm) will be cored with a 1 cm diameter increment corer rather than by excavation. Samples will be frozen on dry ice for transport to the laboratory, and subsequently stored at -80° C until they are lyophilized (Andersen et al. 1991). Freeze-dried samples will then be ground and analyzed for starch and TNC content (Wilson et al. 1995).

Root samples will initially be collected from a subsample of trees at 6-week intervals to identify the appropriate sampling times, e.g., sample periods when tree-to-tree variability is minimal (seasonally synchronous). After initial studies, samples will be taken on a larger number of trees and samples will likely be restricted to late fall after the first frost, early spring prior to bud flush, and mid summer, after full shoot expansion. Additional sample periods may be

necessary depending on the results of the initial studies.

Task 6. Phenology

Subtask 6a: Fine root dynamics

Objectives and background

The objective of this subtask is to employ minirhizotrons to track individual roots or root segments over their lifespan to determine (1) optimum sampling frequency for quantifying fine root dynamics and (2) fine root production and mortality over time and environmental gradients. Data on fine root dynamics will also contribute to the parameterization of the MBL-GEM.

Under normal conditions, the allocation of photosynthate to the various plant compartments (leaves, stems, roots, reproductive tissues) proceeds at set rates, priorities, and seasons. If plants are stressed at a level beyond which the plant can accommodate, this pattern of carbohydrate allocation is altered. It has been shown that ozone exposure causes carbohydrates usually destined for both fine root growth and large root storage to be retained in the leaf instead for maintenance and repair (Hogsett et al. 1985, Andersen et al. 1991, 1997b). Stressors also impact C allocation to fine roots leading to alterations in fine root growth and life span. Carbon transport to roots and new root growth is reduced in ozone treated ponderosa pine seedlings (Andersen and Rygiewicz 1995; Andersen et al. 1991, 1997b). Although we have not yet tested the hypothesis that ozone increases root mortality and turnover, soil organic matter is greater in ozone than control treatments, which is consistent with this hypothesis. In contrast, elevated CO_2 increases carbon allocation below ground resulting in increased fine root area density (Tingey et al. 1996) and fine root life span (Tingey et al. 1997). This sensitivity of fine roots to various anthropogenic stressors suggests that fine root dynamics have the potential to serve as early warning indicators of forest condition.

In most plant communities, root systems are poorly understood and data from natural plant communities are limited (Hendrick and Pregitzer 1996). Minirhizotrons are well suited to study natural plant communities as they are relatively small and permit the continuous, *in situ*, non-destructive monitoring of fine root processes over long time periods. A unique strength of the minirhizotron approach is the ability to study individual roots or root segments over their life-spans. As individual roots are tracked the fine root production and fine root mortality is directly observed. Processes like root production, elongation and mortality are measured separately rather than inferred indirectly using mass-balance procedures on soil cores (Hendrick and Pregitzer 1996). Only a few studies have directly measured root production and mortality as istinct processes which is a significant limitation in forest ecological studies (Hendrick and Pregitzer 1996). Although minirhizotron systems are useful for monitoring fine root production and turnover. they are difficult to use for estimating root biomass (Upchurch 1987, Merrill and Upchurch 1994). However, minirhizotron data have been converted to root length densities which can be used to estimate biomass if specific root length is

known (Upchurch 1987, Merrill and Upchurch 1994).

Methods

Basic minirhizotron approach

Minirhizotron tubes (5 cm inside diameter; fitted with a water-tight PVC plug on the soil end) will be installed at an angle 45° from vertical; extending 1.4 m into the soil (1 m vertical depth). The aboveground portion of each tube is painted to exclude light and covered with a closed-cell foam rubber and a PVC cap to keep out moisture and minimize heat exchange between the tube and the air. The application of minirhizotron technology has been described in detail by Brown and Upchurch (1987). Minirhizotron images will be recorded periodically on S-VHS tape using a minirhizotron camera (Bartz Technology Company, 650 Aurora Ave., Santa Barbara, CA, U.S.A. 93109) (Tingey et al. 1995, 1996). The camera is remote focusing, with a white light source and equipped with an indexing handle that locks into position in an index hole in each minirhizotron tube. The indexing handle (Johnson et al. 1997) has a ratchet advancing mechanism and regularly spaced detents to reliably advance the camera from one field of view (frame) to the next. The indexing handle system insures that the camera is returned to the same position in each tube and travels along the same viewing line each time images are collected. Root images will be recorded on the uppermost surface of the minirhizotron tubes beginning at the bottom of the tubes. In this application the minirhizotron camera has a field of view of about 1.8 cm^2 (1.1 cm high by 1.6 cm wide). Based on tube length and video camera field of view a continuous soil strip representing about 10% of the total surface area of a minirhizotron tube is sampled.

The minirhizotron video images will be analyzed using "MSU-ROOTS", an interactive PC-based software program (Hendrick and Pregitzer 1992, 1996). For analysis, the video images are displayed on a video monitor; the combination of image collection and display on the video monitor magnifies the images approximately 25 times. The software allows the user to review all images and use a mouse to trace various root features (length and diameter) and annotate mycorrhizae and fungal hyphae occurrence. For roots which branch, each branch is tracked as a separate root segment. Each digitized root is assigned a developmental class (SOP 6.02, *Minirhizotron Image Data Extraction Using the MSU-ROOTS Software System*) All roots are "new" at the first observation while roots that disappear between samplings are classified as "missing. Roots are separated into coarse (> 2 mm diameter) and two classes of fine roots ($\leq$ 1 mm and 1 to $\leq$ 2 mm).

The occurrence of mycorrhizal fungi are inferred by the presence of monopodal, bifurcated or highly branched root tips. Fungal hyphae will also be noted but it is not possible to distinguish among various types of fungi from the hyphae observed in the images.

Several types of data will be obtained from the minirhizotron images (Hendrick and Pregitzer 1992, Majdi 1996): (1) occurrence of fine ($\leq$ 2 mm diameter) roots, ectomycorrhizae, and fungal hyphae, measured as the proportion of images

containing these structures (Tingey et al. 1995, 1996); (2) length and diameter of individual fine root segments (Tingey et al. 1996), and (3) lifespan and mortality of individual fine root segments and mycorrhizae (Johnson et al. 1997; Rygiewicz et al. 1997) (see also Subtask 9a below).

Sample frequency to assess fine root production and mortality

Minirhizotrons are well suited for measuring fine root production and mortality (e.g., Hendrick and Pregitzer 1966, Majdi 1996). However, the appropriate sample frequency has not been determined and the frequency may vary among species. To estimate total fine root production it is important not to "miss" any production during the interval between image collections. This is especially a problem if new roots are produced, die and disappear between image collection intervals. In ponderosa pine, median lifespan of fine roots can be as short as 50 days during the summer when the soils are warm (Johnson et al. 1997). Given such a short median lifespan for ponderosa pine and assuming that the median lifespan for Douglas-fir is similar, it will be necessary to collect images every 2 to 4 weeks to estimate the production and mortality rates reliably for Douglas fir. The key feature of this research is to determine the best sampling frequency to determine "total" fine root production.

We propose to place minirhizotron tubes around several Douglas-fir trees and collect images at least twice a week for a year. The images will be digitized and then the data base subsampled at different time intervals to establish the minimum sample frequency required to capture fine root production and mortality. Soil temperature and soil moisture will be measured over the course of the experiment. Air temperature, solar radiation and precipitation are available from an adjacent meteorological tower.

Field measurement of fine root production and mortality

Little data exist showing the total production of fine roots and their mortality rates, under natural conditions, at the stand level. The sample collection frequency will be based on the previous study that established the optimum frequency to quantify all new fine root production. To determine fine root production and mortality rates, minirhizotron tubes (25/site) will be placed at the High and Low intensive field sites. The minirhizotron tubes will be co-located with litterfall traps (section 3.2.1), N-mineralization studies (section 3.2.1), and soil cores collected to estimate fine root standing crop (subtask 4a). As the various samples are co-located we will compare various methods of estimating new root production and turn-over (section 3.2.1).

The data on fine root production and mortality will also be used to determine seasonal patterns of fine root formation and loss for the forest stands. These data will be linked with seasonal stem growth patterns obtained from dendrometers (see section 3.2.1) placed on individual trees (covering the species present) at each site to determine the temporal relationship between fine root dynamics and stem growth.

3.3.2. Resource utilization-consumers

Task 7: C and N Transformations

Subtask 7a: Soil organic matter fractions

Objectives and background

The goal of this subtask is to characterize the forms and chemistry of C fractions in a range of forested soils. Comparisons between the soils at the intensive forest sites (section 3.1) and the adjacent clear-cuts will be used to identify changes in SOM (O horizons and mineral soil) due to management and to develop SOM-based indicators. Soils are an important component of forested ecosystems having vital roles in supplying water and nutrients for plant growth and maintenance and in ecosystem stability and productivity (Ågren et al. 1996). Forests generally have a fairly tight nutrient cycle that relies upon decomposition of plant derived organic matter to supply nutrients. Perturbations in decomposition that upset or interrupt the nutrient cycle can affect the stability of the system. Stressors such as elevated CO_2 and/or management may affect soil processes, such as decomposition, to that extent that the whole system is affected. The character and composition of SOM fractions may provide valuable information about the system stability and the effects of stressors on the trajectory of system stability (e.g., decreasing SOM may signal decreasing system stability).

The amount of soil organic matter in any soil is a function of: climate, relief, parent material, time, and quantity and quality of organic inputs. Natural and anthropogenic factors that affect any of these SOM forming factors will affect the quantity and quality of SOM (Oades 1988). Because SOM can have mean residence times of up to hundreds to thousands of years (Post et al. 1982), its characterization may provide long-lived metrics of ecosystem status and indicators of ecosystem change.

Methods

Basic soil and litter chemistry

Litter and mineral soil samples will be collected with a corer four times per year, to capture seasonal variability, in the forest and adjacent clearcut at the intensive (Falls Creek and Toad Creek Road) field sites. Samples will be collected from five randomly selected subplots of the intensive field sites. Soil samples will be collected from all horizons. Litter layer samples will be separated into two horizons, the Oi (slightly decomposed) and Oa (highly decomposed). Each time samples are collected their bulk density and moisture content will be measured.

The concentrations of nutrients will be measured in the soil and litter samples. Field moist samples will be used for pH, extractable N and extractable cations (Nutrient Analysis SOP#3.02 *Macro-nutrient analysis by suppressed ion chromatography with conductivity detection;* Alpkem Autoanalyzer SOP# 3.10; and ICP Analysis SOP#3.04 version 2.00). Extractable S and P will be run on fresh mineral soils (Soil, Litter and Plant Tissue Preparation EP#01). Freeze-dried samples will be used for total C and N (Carbon and Nitrogen Elemental Analysis SOP#3.01), and a total elemental analysis (ICP Analysis SOP#3.04 version 2 00).

Ratios of SOM to various nutrients may provide useful system indicators.

Soil organic matter quality

For this study quality refers to the ease with which SOM is decomposed. Rapidly decomposed SOM serves as a desirable substrate for decomposers and is high quality. Recalcitrant SOM is an undesirable substrate and consequently poor quality, and more likely to have a much longer residence time.

Samples from the uppermost soil and litter collected as described above will be used for the studies described below. We propose using two measures of soil and litter quality. One will be based upon the methods given in Cambardella and Elliott (1992) who developed a physical separation method that links measurable fractions of SOM to kinetically-defined pools. This method includes dispersion, size fractionation, density separation and chemical analysis. Second we will use incubation studies to characterize the CO_2 production, O_2 consumption, N-mineralization and decomposition of soil and litter samples (Nadelhoffer 1990; Carnicala et al. 1995). Soil and litter samples will be incubated under optimal moisture and temperature conditions to facilitate rapid decomposition. During the incubation, production of CO_2 and consumption of O_2 will be measured. These data will be used to investigate the kinetics of decomposition. Also, during the incubations the soil and litter samples will be subsampled and total C, N and extractable N will be measured. These data will provide information about changes in the quantity of soil and litter C and N pool and the timing of N mineralization. These studies will provide information on the spatial and temporal differences in SOM quality and the differences due to management (e.g., cut versus uncut). This information will lend itself to the development of SOM-based indicators of forest condition.

Soil organic matter characterization

Classical SOM characterization methods use assorted extracting solutions to separate it into operationally defined organic fractions (e.g., base extractable humic acids, Stevenson et al 1989). We propose using solid-state cross-polarization/magic-angle-spinning ^{13}C NMR (CP/MAS ^{13}C NMR) to characterize the composition of SOM. This technique has been described by Baldock et al. (1992) and can be used to examine the composition of SOM. Because the CP/MAS ^{13}C NMR technique can be used to observe subtle changes in the composition of SOM we will use this technique to characterize the C in a subset of the soil and litter samples; if the results appear to be promising more samples will be analyzed. Instrument time permitting, we intend to integrate CP/MAS ^{13}C NMR characterization with our incubation studies and with the SOM separation methods described by Cambardella and Elliott (1992 and 1994) and by Baldock et al. (1992). These results will provide information on SOM processing. Other techniques, such as those described by Ryan et al. (1990) may also be employed to facilitate integrating this SOM research with the MBL-GEM component (section 3.2.1).

Task 8: C and N Losses

Subtask 8a: Efflux of soil carbon

Objectives and background

The specific research objectives of this subtask are to: (1) quantify the diurnal and seasonal changes in soil C efflux, as CO_2, using a newly-developed surface *in situ* measurement device; (2) quantify the seasonal changes in the relative contributions of root, litter decomposition, and SOM oxidation to total soil efflux; (3) calculate the direction in the net change of SOM; and (4) use output from these results to contribute to the evaluation and verification of the MBL-GEM (section 3.2.1). The soil-efflux fractionation method and a newly-developed technique for automatically and semi-continuously monitoring soil efflux will be used to address these objectives.

The direction of change in long-term functioning of a terrestrial ecosystem, especially of temperate forests, relies partly on changes in the amount of soil organic matter (SOM) (Tilman 1997). Specifically, forest functioning is related to the balance between accrual of SOM when the system exists either in the undisturbed condition or while it is recovering from a disturbance, relative to SOM losses due to disturbance. If the system can not replenish lost SOM before disturbance reoccurs, the trajectory is negative. Similarly, if a chronic stress causes SOM to be depleted faster than it can be replenished, ecosystem health may be compromised. Conversely, if the chronic stress depletes SOM at a rate less than the SOM replenishment rate, long-term functioning may not be affected.

Observing a net change in SOM can be difficult because it may entail measuring small changes in a large pool. In our work in the teracosms on the effects of elevated CO_2 and climate change on a Douglas-fir/soil system, we developed a method to partition soil litter CO_2 efflux into its component sources of root respiration, litter decomposition and oxidation of SOM (Lin et al. 1998). This method relies on the simultaneous determination of the natural abundance of ^{13}C and/or ^{18}O in the soil efflux, and of the C and water in the litter and soil. By using a mixing model, soil CO_2 efflux is partitioned into its components by using a 2-end-member linear model for the $\delta^{18}O$ value of CO_2, and a 3-end-member triangular model for the $\delta^{13}C$ value of the efflux (Lin et al., 1998). Thus, efflux of C from the SOM pool can be determined. Net flux (influx - efflux) of SOM C is calculated after determining influx of C to the SOM pool using a similar approach relying on ^{15}N and ^{13}C (analyses and model development are on-going for the work being done in the teracosms).

Methods

Determining the relative contributions of each C source (root, litter decomposition, and SOM oxidation) to total soil efflux involves the following two experimental techniques: 1) quantifying and capturing soil CO_2 efflux for subsequent isotopic analysis, and 2) collecting and processing soil, root, and soil water samples for isotopic analysis. Details for the method to determine influx of C to the SOM pool using ^{15}N and ^{13}C are still under evaluation from samples collected in the TERA 1 experiment.

Soil CO_2 efflux estimation

Soil efflux will be estimated using a newly-developed technique for automatically and semi-continuously monitoring soil efflux. The linear *in situ* soil-litter respirometer (LISR) was developed to estimate soil efflux automatically, semi-continuously, and non-destructively. The sample area of a single LISR is 1 cm wide by 1 m long, thereby providing an integrated measure of soil efflux. Efflux estimates are recorded automatically and semi-continuously using a Campbell Scientific CR10X datalogger. Placing the LISR on the soil-litter surface minimizes disturbing roots or mycorrhizal hyphae located in the litter layer or mineral soil.

Prior to using the LISR we will evaluate it under field conditions to see if it has a "footprint" effect that could bias the results of CO_2 release rates. At present, the relative and absolute accuracy and precision of soil efflux estimates obtained by the LISR and the LICOR Soil Respiration Chamber are being evaluated using a controlled-environment test chamber. Soil efflux estimates derived by the LISR are within 5% of values obtained using the LICOR headspace chamber. Low labor requirements for its operation, and high portability make the LISR a useful tool for measuring soil efflux in a variety of ecosystems. Assessing spatial variability in soil efflux is accomplished by hooking several LISRs to the datalogger equipment. Automated operation of the LISR readily permits collection of data describing the diurnal and seasonal patterns in soil efflux. The LISR can be modified to readily permit soil gas samples to be collected for stable isotope analysis.

A single estimate of soil efflux will be estimated once every hour at five sampling locations over a 48-hour period. These estimates will be made once every four weeks for an entire calendar year (i.e., 13 sample periods) at the intensive (Falls Creek and Toad Creek Road) field sites. The five sampling locations will be positioned adjacent to any litterfall traps, nitrogen mineralization tubes, minirhizotron access tube locations, and soil moisture and soil temperature sensors installed so that soil processes can be more fully described and incorporated into any statistical inferences made. We will also attempt to measure wintertime soil efflux, thereby more fully understanding the contribution of soil processes to the annual ecosystem C cycle. Prior to snowfall, five LISRs will be placed on the ground and their gas sample plumbing lengthened for wintertime measurements. At each sample period when snow is present, another LISR will be placed directly over the LISR positioned on the ground. This will allow near-simultaneous estimates of soil efflux at the soil surface and at the snow-atmosphere interface.

Soil respiration partitioning

The dual isotope (^{18}O and ^{13}C) method is based on the differences in natural abundance of ^{13}C among the three C sources, and differences in natural abundance of ^{18}O between litter water and soil water. We demonstrated the success of the method in the TERA 1 experiment where the source of CO_2 for the newly-formed C (i.e., C accrued during the climate exposure treatments) was a tank CO_2 of geologic origin (i.e., highly depleted of ^{13}C). We expect that the differences in natural

abundance of the isotopes among the sources of soil efflux at our field sites will be sufficient so that the method will work. It is the differences in the respective $\delta^{13}C$ and $\delta^{18}O$ values among the component pools for the two isotopes that will determine if the method will work. For example, in the field, the $\delta^{13}C$ value now for recently-fixed CO_2 is likely not too different from that of CO_2 fixed in the past. From our preliminary data of respective differences in the field, there are sometimes sufficient differences which are dependent on climatic conditions. We will undertake an expanded analysis of this work initially to confirm our expectations.

Although the use of stable isotopes of C and O individually has been used successfully to measure a variety of ecosystem processes (cf., Keeling et al. 1979, Ehleringer et al. 1986, Peterson and Fry 1987, Farquhar et al. 1993), the simultaneous application of these isotopes to study root vs. microbial fractionation has not been accomplished until recently (Lin et al. 1998). The relative contributions of root and microbial respiration to total soil efflux will be estimated by measuring the ratios [R] of $^{13}C/^{12}C$ and $^{18}O/^{16}O$ in roots, soil water, and forest litter. These isotopic ratios are usually expressed in δ units relative to a standard [Pee Dee Belemnite or Standard Mean Ocean Water (SMOW)] where $\delta^{13}C$ or $\delta^{18}O = ((R_{sample}/R_{standard})-1)1000$.

The C isotopic ratio of soil efflux is determined by the $\delta^{13}C$ of its C sources which are derived primarily from roots and litter. Litter, comprised of tissue with C isotopic ratios reflecting initial fixation of atmospheric CO_2 and re-fixation of soil-respired CO_2 (soil efflux). Therefore, a C isotopic ratio gradient exists, vertically and horizontally in the forest canopy. An accurate assessment of the $\delta^{13}C$ across the presumed vertical gradient of $\delta^{13}C$ in the litter needs to be made and a representative value needs to be determined. It is assumed that the vertical gradient of litter $\delta^{13}C$ also exists due to seasonal variation in canopy photosynthesis. Because root respiration is not subject to these seasonal re-fixation processes, its $\delta^{13}C$ should be different from the litter.

Surface $\delta^{18}O$ values of soil water are higher than values deeper in the soil profile due to a higher evaporation rate of the lighter oxygen isotope (Craig and Gordon 1965). As CO_2 is produced, the oxygen isotope of CO_2 comes into equilibrium with the oxygen isotope of the soil water at the site of CO_2 production. In other words, the isotopic ratio of soil efflux depends on the $\delta^{18}O$ value of the soil water at the site where the CO_2 is produced since soil efflux quickly comes into equilibrium with soil water through a carbonic anhydrase-mediated reaction (Hesterberg and Siegenthaler 1991). Consequently, one is able to distinguish whether the C in the soil efflux originated from SOM oxidation or from litter decomposition.

The following sampling protocol will be followed:

Air: Soil efflux will be collected by shunting gas collected by a LISR into a 2 liter vessel. A single sample will be collected from each LISR once during each sample period (n = 13). Respired CO_2 gas samples will be collected at approximately

the same time as soil, root and litter; and soil H_2O samples are collected (see below). The soil efflux sample will be cryogenically extracted and then the isotopes analyzed using WED's Finnigan delta S Isotope Ratio Mass Spectometer (IRMS). It is critical to extract the C from the CO_2 within 4 hours of collection to avoid altering the C isotope signature through reactions with the water vapor present in the gas sample.

Soil, roots, and litter: Three soil cores, 2.54 cm diameter, will be collected within 5 cm of each of five LISR's during each sampling period (n=13). Soil cores will be collected at equal intervals (i.e., 25, 50, and 75 cm) along the 1 m long LISR once each sample period. Soil cores will be collected at approximately the same time that soil efflux samples are collected. All soil cores will be separated into litter, and if possible, shallow (A-horizon) and deep root zone (B- and C-horizons) components and the components placed in a dry ice container for transport to WED. Each soil component (A, B, and C horizons) will be further divided into root and soil fractions at WED.

A 50 mg sub-sample from the shallow (A horizon) and deep (B and C horizon) root zone soil sample will be dried and ground for isotopic analysis. $\delta^{13}C$ values for each component will be determined by analysis in the WED IRMS. Water samples from litter, and shallow and deep roots will be cryogenically extracted using a vacuum line extractor. A 1 cc water sample of each component will also be analyzed for isotopic ratios, using a modified CO_2 equilibrium method described by Socki et al. (1992), and the WED IRMS.

Task 9: Complexity and Function of the Soil Food Web

Subtask 9a. Mycorrhizal symbionts

Objectives and background

The main objectives of this task are to: (1) calculate lifetimes of mycorrhizae. i.e., time between formation and disappearance to determine turnover, (2) measure carbon allocation to extraradical hyphae, (3) calculate colonization levels of root tips (% mycorrhizal tips), and 4) estimate natural variation in the community of fungi forming mycorrhizae. Mycorrhizae, by acquiring nutrients for their hosts and acting as carbon sinks, are important for plant carbon allocation (Rygiewicz and Andersen 1994), plant nutritional status (Rygiewicz et al. 1984a, b; Bledsoe and Rygiewicz 1986), nutrient cycling (Harley and Smith 1983), and the regeneration of plants and, hence, the sustainability of forest ecosystems (Perry et al. 1989). We propose therefore that measures of the structure and process rates of the mycorrhizal community have the potential to serve as forest indicators.

Mycorrhizal fungi are among the first soil biota to receive C from plants, and to acquire nutrients and water taken up by plants. Consequently, subsequent release of root/mycorrhizal C as exudates and turnover, supports soil foodweb activity and many "downstream" processes in terrestrial ecosystems. Process rates among mycorrhizae and mycorrhizal fungi are highly variable *in vitro*, and also presumably in the field (e.g., Harley and Smith 1983, Zhu et al. 1988, Wagner et al. 1989). It is important, therefore, to evaluate rates while considering the

community structure of the fungi forming the symbiosis. Stressors may influence, directly or indirectly, the capabilities of mycorrhizal fungi to colonize roots, and acquire nutrients and other resources. (Andersen and Rygiewicz 1991). When scaled to the ecosystem, changes in the community structure of mycorrhizae may greatly alter resource balances. For example, Fogel and Hunt (1983) found that up to 70% of net primary productivity in a Douglas-fir ecosystem was invested in growth and maintenance of roots and mycorrhizae.

How anthropogenic stressors acting on forest ecosystems affect community structure and processes of mycorrhizal fungi is not well known. Stressors due to air pollution or global change can increase, decrease or have no effect on C allocation belowground with subsequent similar changes in mycorrhizal fungi (Andersen and Rygiewicz 1991, 1995). Responses to stressors may result in larger or smaller root systems with different allometric relationships among root sizes (e.g., changed proportions of mycorrhizae, and non-mycorrhizal fine, intermediate and coarse roots). Changes in C allocated belowground also may cause changes in colonization rates by mycorrhizal fungi and development of extraradical hyphae. Considering the potential variability in C requirements of fungal species, it is prudent to consider both the dynamics and demographics of mycorrhizal fungi in ecosystem indicator development.

Methods

Lifetimes, carbon allocation to extraradical hyphae, and colonization levels

These will be determined using the same minirhizotron tubes described in subtask 6b. The dates of formation and disappearance of each mycorrhizal tip encountered in the tubes will be recorded and used to calculate lifetimes. If a large enough sample population is encountered, cohort analyses (monthly, seasonal, annual, etc.) for lifetimes will be done. Total number of non-mycorrhizal and mycorrhizal root tips will be recorded to calculate colonization rates. For each minirhizotron frame containing mycorrhizae, an estimate will be made of the percent of the screen containing hyphae that appear in some way associated with a mycorrhiza. This is an imperfect approach, but it will allow us to estimate C allocation to extraradical hyphae *in situ*. Independent estimates of mycorrhizal colonization will be determined using root samples obtained from the soil cores taken periodically from the intensive field sites in coordination with the sampling scheme described in subtask 4a and 6.

Natural variation in community structure of fungi forming mycorrhizae

The structure of the mycorrhizal fungal community will be assessed at the individual mycorrhizal tip level. Soil cores will be taken periodically from the intensive field sites in coordination with the sampling scheme described above (Subtask 4a and 6b). Results from the TERA I project indicate that overall seasonal variation in the community

structure of mycorrhizae on Douglas-fir is low (i.e., comparing spring samples, taken when soils are at field moisture capacity and cool, with fall samples, taken when soil moisture is very low and temperatures are high). We do not have seasonal results for a more mature forest stand, so we are unable to suggest an exact sampling schedule. We intend to sample more frequently during the first year, and then adjust the coring schedule accordingly.

The structure of the mycorrhizal community will be assessed both phenotypically (categorizing mycorrhizae into morphotypes based on gross morphology), and genotypically (using molecular methods). We have become proficient at categorizing mycorrhizal tips of Douglas-fir seedlings of the TERA 1 project. After the tips are assigned to morphotypes, a selection of replicate tips per morphotype is subjected to polymerase chain reaction (PCR)-RFLP analysis to determine the fidelity of the initial categorizations. Additional PCR-RFLPs are produced in any morphotype where genetic variation occurs. If necessary, the entire morphotype community is then re-categorized according to the molecular analyses. Ontogenetic processes and certain stressors (such as N loading) may interfere with using only gross morphological features to analyze the community since morphology may be affected.

We found that the ribosomal DNA primer pair generally recommended and used to produce PCR-RFLPs of fungal DNA from ectomycorrhizae does not work for Douglas-fir (both the host and mycobiont DNA are amplified). We identified other ribosomal DNA sequences having high homology with basidiomycetous DNA to construct primers for Douglas-fir mycorrhizae. We constructed two nested primer pairs which are used in nested-PCR amplifications. Using two primer pairs, rather than one pair, increases the specificity for the mycobiont DNA. We tested the two new primer pairs with a limited number of Douglas-fir mycorrhizae and obtained mycobiont amplification without host DNA amplification. The PCR reaction is being optimized.

Until we can estimate natural variation in the mycorrhizal community structure, temporally and spatially, we will not be able to distinguish natural cycles due to season or succession from changes induced by anthropogenic stressors. The work proposed here in this task, along with work completed or ongoing in the TERA 1 and TERA II experiments, will yield an analysis of how the host-symbiont association integrates changes in C and N allocation belowground. Changes in community structure, coupled with changes in other measures developed in this plan may prove useful as indicators.

Subtask 9b. Soil foodweb biota

Objectives and background

The objectives of this sub-task are to (1) investigate the composition and complexity of soil foodweb organisms including microarthropods, nematodes, bacteria, and fungi present in litter, soil, and the rhizosphere as a function of forest successional state and season, (2) measure the molecular diversity and activity of associative N_2-fixing bacteria, fluorescent pseudomonads, and symbiotic and free

living fungi, and (3) measure key physiological and metabolic soil microbial community activities associated with the above parameters. To address these objectives, we are taking a diverse methodological approach. This involves the use of established techniques (population biology) as well as the application of newer methodologies such as community metabolic analyses (Biolog) and DNA fingerprinting techniques to relate foodweb composition to important functional aspects of forest responses to stressors. We propose that these structure and functionality parameters have the potential to serve as indicators of C and N processing in forest soils.

The quantity and quality of herbaceous and woody plant materials and litter may be expected to vary with different environmental and edaphic conditions, plant community composition, successional stage, and anthropogenic stressors. We hypothesize that these variations in organic inputs in turn will affect the diversity, population sizes, and rates of nutrient and C utilization by litter and soil foodweb biota, and hence the rate of C and N cycling (Fig. 3-6). The model shows the linkages between initial fragmentation, detritivores, and trophic roles of invertebrates with other foodweb microbiota and plant tissues, with the subsequent metabolic roles of microbes (bacteria and fungi), in ecosystem level processing of aboveground and belowground plant inputs.

We assume that plants exposed to anthropogenic stressors will have altered patterns of C and N allocation. Differences in the composition of living plant tissues (shoots and roots), litterfall and root exudates will bring about changes in the population size, diversity and functions of foodweb biota in litter and soil. Changes in the population size, diversity and biomass of foodweb components are proposed to affect the rates, amounts and types of microbial metabolism of both C and N compounds. Resultant differences in C and N metabolism will then impact the amount of CO_2 released from soil to the atmosphere and the amount of N available for plant uptake. The amounts of CO_2 released and N available for plant uptake would in turn be expected to potentially impact plant productivity for example. Furthermore, once altered, the foodweb microbiota may induce a feedback that could further affect the nutritional status of the plant/rhizosphere/soil complex.

Methods

Samples will be collected from the intensive and extensive field sites described above (Section 3.1). Samples will be collected from 2 m x 2 m subplots. Litter will first be removed, using gloved hands to prevent cross contamination, from a 20-cm diameter circular area for the forested plots and a 60-cm circle for clear-cut plots to have sufficient biomass. Soil samples will then be taken with 3-cm diameter aluminum cores to depths of 0-10 cm and 10-20 cm; this is where the highest densities of foodweb populations are located. Contents of the cores will be separated into the two depths, mixed by hand, and subsamples taken to satisfy all the biological measurements described

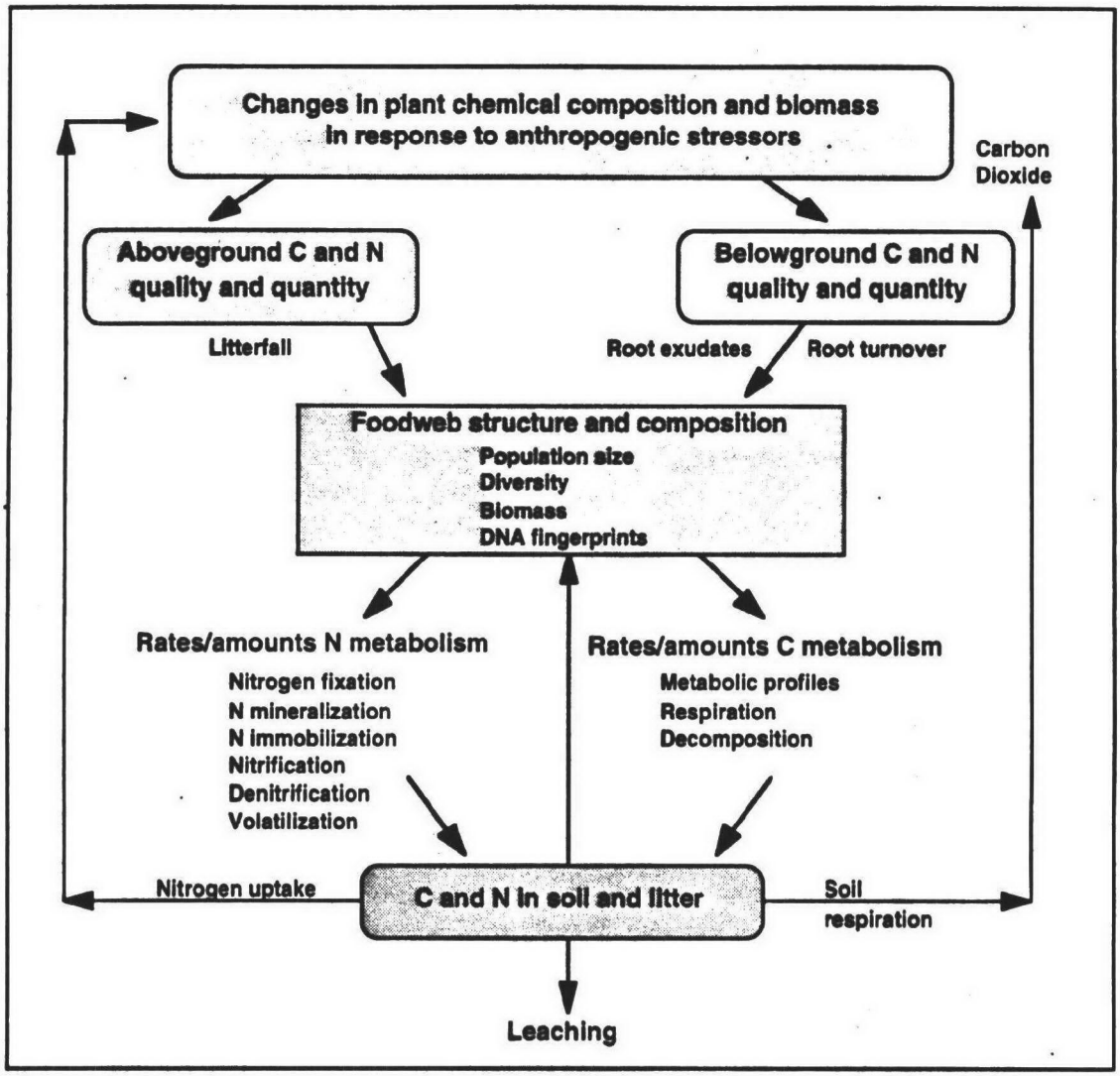

Figure 3-6. Conceptual model showing the role of the soil foodweb in mediating soil C and N metabolism responses to anthropogenic stressors.

below. Over time, samples will be taken diagonally across the 4 m^2 subplots to maximize the space available for coring within the designated plots. Data on soil foodweb structure and composition will be correlated with several of the field measures gathered from the work described above (section 3.2.1, Task 1) or from additional measures described here.

Composition and complexity of foodweb organisms:

Bacterial and fungal counts

Samples of litter and soil will be diluted in sterile buffer, shaken, and spread plated onto selective media as described in Donegan et al. (1995) to determine viable populations of bacteria, fungi, and spore-forming bacteria (USEPA SOP 7.16m *Microbial Plating*).

Fungi found to increase or decrease with the seasons will be grown in pure culture for subsequent taxonomic, phylogenetic and functional characterization based on microscopic morphology (Raper and Fennell 1965, Domsch et al. 1980, Stone 1993, USEPA SOP 7.14 *Fungal Colony Growth and Hyphal Morphology on Agar Media*) and DNA fingerprinting (see below). Colony morphology richness will be determined to identify shifts in clonal and species composition associated with a given season and successional stage. The Shannon-Weaver index will be used to mathematically evaluate fungal richness in the various ecological situations. By using cultural biotyping, colony morphotyping, and DNA fingerprinting patterns we will index fungi so that their potential as indicators may be developed, evaluated, and defined.

Nematode and microarthopod extraction, identification, and enumeration

Studies in a diverse range of soils have shown that soil fauna (nematodes, microarthropods and also protozoa) mediate approximately 15% of the C and 30% of the N turnover in the soil (Anderson 1995). Because nematodes and microarthropods have high species richness and a complete representation of trophic groups (fungivores, bacterivores, herbivores, omnivores, and predators), measuring their numbers and species can provide sensitive detection of changes in many processes of forest ecosystems. Changes in the population dynamics and structure of nematodes and microarthropods have been used as bioindicators of the effects of several environmental stressors (Parmelee 1995, Larink 1997, Zunke and Perry 1997).

To determine the composition and population complexity of soil and litter invertebrates, samples of soil and litter will be weighed and placed into modified Baermann funnels and extracted as described by Donegan et al. (1997) (USEPA SOP 7.04). Total numbers of nematodes will be counted and expressed as the number per g of dry weight of soil or litter. For extraction of microarthropods, soil and litter samples will be placed into modified Tullgren funnels in beakers containing water (Donegan et al. 1997). Microarthropods will be collected and identified to class (Diploploda, Chilopoda, or Symphytla) or to order (Acari, Protura, Collembola, Diiplura, and order of insect) according to Borror et al. (1992). The total number of each class or order will be expressed as the number per g dry weight of soil or litter. In key samples, about 30-50 nematodes per sample will be randomly picked and transferred to slides for identification to trophic groups according to Yeates et al. (1993) and Donegan et al. (1997).

Molecular measurements

Structural and functional analyses of associative nitrogen fixers

Use of DNA oligonucleotides as markers is facilitating *in situ* detection, enumeration, and determination of metabolic activities of both culturable and non-culturable microbes found in natural habitats (Seidler and Fredrickson 1995, Ueda et al 1995). Using modified *nifH* primer sequences, we have found DNA fingerprints that indicate that certain N_2-fixing organisms are unique

to litter in mature forests, but are not detectable in litter from nearby clearcuts (Widmer et al. 1997a). We will use the *nifH* primer to document the kinds and molecular phylogenies of organisms present in the litter and belowground that contribute to N cycling in forests. Acetylene reduction measurements will estimate amounts of N fixed and provide functional evidence to support the molecular studies on *nifH* biodiversity.

DNA fingerprint for N fixation

Information identifying the molecular heterogeneity and phylogenies of N_2-fixing organisms will be achieved by polymerase chain reaction amplification (PCR) of the *nif*H gene following extraction of bulk DNA from litter and soil using methods that we have developed previously (Porteous et al. 1994, Porteous et al. 1997, Widmer et al. 1996, USEPA SOP 7.15m *Bulk Soil DNA*). The *nif*H gene codes for a subunit of the dimeric nitrogenase enzyme and its presence is associated with N_2-fixing activity. Primers for PCR amplification have been synthesized and function well in detecting all phylogenetic groups of N_2-fixing microorganisms. The reaction conditions for amplification have been described (Widmer et al. 1997a). Purified plasmid libraries of characteristic *nifH* PCR products from selected field samples will be constructed and transformed into *Escherichia coli*. Plasmid DNA extracts from the *E. coli* strains will be characterized to validate RFLP patterns and representative clones for each pattern will be subjected to sequence analysis by the Center of Gene Research and Biotechnology, Oregon State University. Phylogenetic analyses of these sequences will be conducted by alignment with those retrieved from Gen Bank using the multiple alignment routine of MacDNASIS pro v3.2. Further details for cluster analysis and construction of phylogenetic trees have been described (Ueda et al. 1995, Widmer et al. 1997a).

Associative N_2- fixation through acetylene reduction

To follow both spatially and temporally the influence of seasons and litter quantity and quality on the kinetics of N_2-fixation and to supplement molecular characterization of *nif*H populations therein, we will measure nitrogenase activity of various types of plant litter. Standard techniques will be used to detect *in situ* associative N_2-fixing activity based on reduction of acetylene to ethylene (Seidler et al. 1972, Bormann et al, 1993).

We will compare rates of acetylene reduction/gm of litter or root for each field plot of interest. Appropriate controls will be included to measure any indigenous ethylene production produced by plant or microbial sources in our samples. Analyses will compare seasons, litter quality and chemistry, meteorological and elevation conditions, and successional stage with the molecular *nifH* patterns and with the kinetics of N_2-fixation at the various sites. A comprehensive assessment of the physical, chemical, and biological associations that impact N_2-fixation in forests will be derived through comparisons with other data gathered in this project.

Use of molecular tools to evaluate phylogeny and diversity of plant-associated Pseudomonas and fungal species

In addition to structural genes such as *nifH*, 16S ribosomal genes have been used by molecular ecologists to study phylogeny and to map the distribution and occurrence of specific bacterial groups through *in situ* analyses. We have developed new DNA primer sequences specific for fluorescent *Pseudomonas* species (Widmer et al. 1997b). Certain strains of this group are rhizobacteria that may promote plant growth, e. g., by inhibiting activities of plant pathogenic fungi. We will use the *Pseudomonas* primers to determine the kinds and distributions of fluorescent *Pseudomonas* species present in litter and rhizosphere samples. In addition, molecular methods for determining the presence, diversity, and functionality of mycorrhizal, saprophytic, and plant pathogenic fungi are available that will be evaluated for their utility to study changes in the litter and rhizosphere of forests.

DNA fingerprint patterns (RFLPs) will be generated from amplified 16S rRNA sequences specific for fluorescent *Pseudomonas* species using bulk DNA obtained from litter, soil, and rhizoplane samples. Amplified 16s rDNA sequences will be digested to generate DNA fingerprints that are unique to the various species of fluorescent pseudomonads (Widmer et al. 1997b). DNA fingerprint patterns from litter will be compared with soil and rhizosphere samples taken directly below the litter. These data will provide new information about the origins of ecologically significant groups of rhizosphere bacteria in forest ecosystems and their response to management. Similar tools will also be developed for fungi (see next paragraph). These results will provide some of the first molecular measurements to document possible ecological linkages between aboveground litter and forest management strategies and belowground plant root colonization by beneficial bacteria. If the populations in litter and soil are consistently different from those on the roots, we will determine whether plant rhizoplane populations are dictated by the plant species, are influenced by aboveground management, or are influenced by other, unidentified processes.

We will also use molecular tools to generate community DNA fingerprints that represent fungal species present in rhizoplane, soil, and litter samples. Primer pairs will be identified, based on small subunit (18S) nuclear rDNA sequences, to obtain community fingerprints that can be used to screen for changes in fungal diversity that occur belowground in forests. We will use the same field samples as described above as sources of DNA. Using standard cloning and sequencing methods as described above, sequence information will be used to identify and elucidate phylogenetic relationships (White et al. 1990, Egger 1994, Edel et al. 1995, Claasen et al. 1996, Perotto et al. 1996, O'Donnell et al. 1997) and functional roles (Kreuzinger et al. 1996, Howe et al. 1997, Perotto et al. 1997) of both free-living and mycorrhizal fungal species. DNA fingerprints will also be generated from isolated pure cultures, most likely primarily of culturable Deuteromycete species, which will be obtained from field samples. This will allow us to ascertain and document the origins of culturable fungi

present in community fingerprint patterns obtained from bulk DNA extracted from environmental samples.

Community metabolic profiling of microbes by the Biolog technique

The Biolog approach (Garland and Mills 1991), initially developed to identify pure cultures of bacteria based on patterns of utilization of 95 different C and N substrates, has recently been successfully used to characterize microbial communities (Ellis et al. 1995, Garland 1996a,1996b, Grayston and Campbell 1996). The technique has also been used to evaluate the effects of land management, agricultural practices, and natural disturbances on microbial communities (Zak et al. 1994, Bossio and Scow 1995, Willig et al. 1996). We will use the Biolog approach to identify potential changes in metabolic profiles or substrate utilization patterns that may occur over time (1) under different land management practices and (2) with plant communities of different successional stages in the Oregon Cascades. The types of changes we hope to find are in substrate utilization profiles of given compounds or groups of compounds (e. g., sugars, sugar alcohols, amino acids, aromatics or xenobiotics), which are represented on the Biolog plates.

Community-level metabolic fingerprints of the rhizosphere microbial communities (Garland, 1996) will be generated using Biolog GN microplates (Biolog, Inc., Hayward, CA) (USEPA SOP 7.17m *Biolog Community Analysis*). Three replicate extracts will be prepared for each sample type, incubated, then read with a microplate reader (Molecular Devices, Inc., Sunnyvale, CA; Di Giovanni et al. 1997). We will treat and process the rhizoplane and rhizosphere samples for bacteria according to the methods of Di Giovanni et al. (1997). Substrate utilization data will be analyzed by principal component analysis (PCA). Analysis of variance of PCA scores will be used to determine whether statistically significant differences occur between samples.

Substrate induced respiration (SIR) /biomass

The size and activity of the soil microbial biomass needs to be measured to fully understand nutrient fluxes and biogeochemical transformations in natural ecosystems (Horwath and Paul 1994). We will measure substrate induced respiration (SIR) on selected samples of litter and soil at strategic sites associated with the other samples being intensively analyzed as described in this subtask (USEPA SOP 7.19m *Substrate Induced Respiration*). Samples will be analyzed with and without glucose additions using standard methods (Anderson and Domsch 1978). We will also conduct SIR analyses with selected antibiotic inhibitors to measure the separate contributions of bacteria and fungi to the total respiration in unique types of litters and soils (Anderson and Domsch 1975, Beare et al. 1990).

4. INTEGRATION WITH OTHER WED PROJECTS

The Forest Ecosystem Indicators Project is one of several in the Terrestrial Plant Ecology Branch of WED, and its coordination with other projects in the Branch greatly enhances its potential for success (Fig. 4-1). The indicator project's

initial emphasis is field research in the Cascade forests, consequently, climatic, edaphic and management practices are the principle stressors for which data will be collected. The ability to test models and indicators against additional anthropogenic stressors of interest to EPA will significantly improve the utility to the Agency of the resultant models and indicators for use as both assessment and predictions tools and increase the reliability of indicators of condition that are developed. Consequently, measures, stress-response functions and models from other research projects being conducted in the Terrestrial Ecology Branch will be used to strengthen the indicator project by providing specific data on the response of trees and forest ecosystems to known exposures of specific stressors (Fig. 4-1).

To investigate how the measures and models developed in the Forest Ecosystem Indicators Project may respond to a range of other environmental stressors (increased temperature, elevated atmospheric CO_2 concentrations, and ozone), we will rely on the integration with three other projects in the branch (Stress response projects--Fig. 4-1). Many of the same measures and models will be investigated by the same scientists in other stress-response experiments.

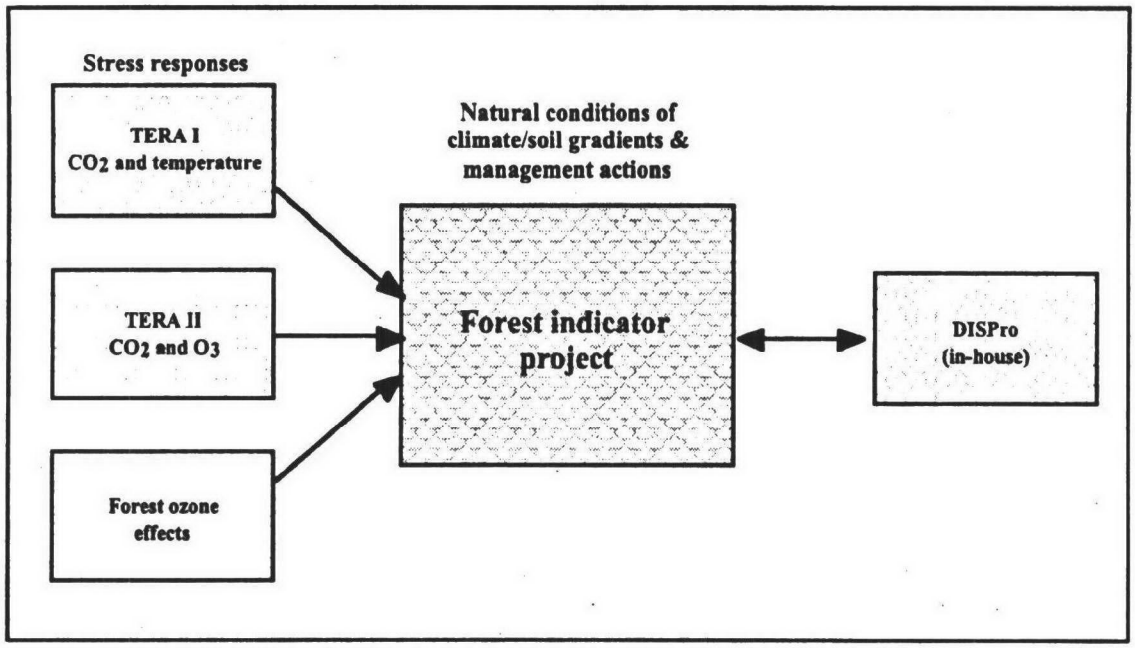

Figure 4-1. Relation between the forest indicator projects and other research projects in the Terrestrial Ecology Branch of WED.

Effects of CO_2 and Climate Change on Forest Trees (TERA I experiment) - The project assessed the effects of elevated CO_2 (ambient and ambient +200 μmol mol^{-1} and temperature (ambient and ambient +4 C) on a Douglas-fir (plant and soil) system over a four-year period. The experimental research was conducted in the Terrestrial Ecophysiological Research Area (TERA) which is a set of sun-lit controlled environment chambers located in Corvallis.

The trees were subjected to the typical wet-dry seasonal soil moisture cycles and relied on soil biological processing of litter for nutrients The experiment was initiated in June 1993 and was completed in August 1997. The effects of elevated CO_2 and temperature were assessed on individual rhizosphere and canopy processes as well as developing system budgets for C and N. System carbon, nitrogen and water fluxes were measured using the mass balance approach -- by accounting for inputs and losses from the terracosms, as well as internal differentiation between plant and litter/soil flux components. Nondestructive measures were made of needle, shoot, and bud development. Minirhizotron tubes and a minirhizotron camera system were used to monitor fine root dynamics and soil cores were collected to follow changes in fine root biomass. Soil and litter samples were also collected to follow soil flora and fauna. A key objective TERA I to collect the necessary data to parameterize TREGRO (an individual plant growth model) and the MBL-General Ecosystem Model [GEM] (an ecosystem level biogeochemical model).

Interactive Effects of O_3 and CO_2 on the Ponderosa Pine Plant/Litter/Soil System (TERA II experiment) - The project will assess the interactive effects of tropospheric ozone (O_3) and elevated (CO_2) on critical indicators of ecosystem function: carbon (C), nitrogen (N), and water (H_2O) cycling. The study will test three hypotheses: (1) elevated O_3 decreases C, N and H_2O cycling rates; (2) elevated CO_2 increases C, N, and decreases H_2O cycling rates; and (3) elevated CO_2 eliminates negative effects of O_3 on C and N cycling rates. The study was initiated in the terracosms in the Spring of 1998 using ponderosa pine (an O_3 sensitive and CO_2 responsive species) seedlings growing in a reconstructed ponderosa pine soil and litter layer from the east side of the Oregon Cascade Mountains. The research is an integrated study with experimental and modeling components. The experimental research addresses: (1) system gas exchange; (2) plant phenology, allometry and carbon allocation; (3) litter and soil/rhizosphere ecology; (4) litter and soil chemical and physical properties; and (5) system materials budgets, pools and fluxes The modeling research will use the MBL-GEM to evaluate effects on C and N cycling, and TREGRO, to study the potential impact of increased O_3 and CO_2 on photosynthesis, respiration, C accumulation, and C allocation. This research will provide unique information on the responses of ecosystem functions related to the interactions of O_3 and CO_2. The research will also provide a complementary set of measurements for many of the parameters (plant, soil, and litter) that will be taken in the field as part of the forest indicators project. Because the measurements in the terracosms will be under clearly defined stress conditions they will highlight the field indicators most useful for detection of environmental stress.

Effects of tropospheric ozone on forest trees (Forest Ozone project) - The project is developing a biological database to support a secondary National Ambient Air Quality Standard (NAAQS) for ozone, which includes: (1) developing an understanding of the nature and extent of ozone's effect on forest trees, and (2) developing a meaningful index of ozone exposure that

can be used to protect forests. The research is conducted in the open-top chamber facility located at Corvallis. The research foci include: (1) seedling/small tree response to ozone in open-top chambers, (2) extrapolation of responses measured in small trees to larger trees, and (3) assessment of ozone exposure in forests across the country and estimating the effects of ozone on those forests. Two important components of the experimental studies on small trees are: (1) physiological and growth responses to multiple stresses (e.g., ozone and nitrogen limitation), and (2) characterizing below ground responses of plants to ozone, including root physiology and rhizosphere activity of free-living organisms. Much of this information is being used directly or indirectly to inform simulations models, in particular TREGRO, as a means of projecting the responses seen in experimental studies across time and space.

The ability to provide stress-response functions and incorporate specific stress data into the models provides a unique advantage that other groups developing ecosystem indicator do not enjoy. This integration of stress-response studies with field studies of climatic and edaphic stressors and management practices is a unique strength of the forest indicator project.

Regional Validation of MBL- (GEM) General Ecosystem Model (Demonstration Index Site Project -DISPro) Project

The primary objective is to develop a parameterization of MBL-GEM that can be used as a risk assessment tool for ecosystems in the Olympic National Park and the Pacific Northwest in general. Specifically, we will use MBL-GEM to:

- Assess and predict future responses of forest ecosystems to natural and anthropogenic stressors, including changes in temperature, precipitation, cloudiness (light), CO_2, ozone, and N deposition

- Link changes in condition to likely stressors

- Identify efficient and sensitive indicators (early warning measures) for loss of ecosystem integrity and sustainability.

Models play a prominent role in ecological risk assessments because they are the primary means for relating stressors to probable effects, and for making meaningful extrapolations across scales of time, space, and biological organization. Models are particularly important for risk assessments at the scale of ecosystems because it is exceedingly difficult to experimentally isolate the interactive effects of natural environmental driving forces (temperature, precipitation, cloudiness, etc.) and anthropogenic stressors (e.g., air pollutants, climate change, land use). Process-based models that simulate biogeochemical cycles or forest succession, for example, can help improve such assessments by providing a self-consistent synthesis of the results of many experiments. The synthesis provided by these models includes the interactions among ecosystem processes that give rise to the synergistic responses to multiple factors. Under DISPro, we propose to use

the Marine Biological Laboratory's General Ecosystem Model (MBL-GEM; Rastetter et al 1991) to assess and predict how natural and anthropogenic stressors may affect the health and sustainability of ecosystems in the Olympic National Park.

The MBL-GEM is intended to be generally applicable to most terrestrial ecosystems and has been used in the past to analyze the biogeochemical responses of temperate deciduous forests, tropical evergreen forests, and arctic tundra to changes in atmospheric CO_2 concentration, N deposition, temperature, irradiance (cloudiness), and soil moisture (Rastetter et al 1991, 1992, 1997; McKane et al. 1995, 1997a, 1997b). In addition, the EPA has recently parameterized MBL-GEM for forest ecosystems located along an elevational gradient in the South Santiam watershed in the western Cascades of Oregon, as a first step toward the development of a regionally robust parameterization for Pacific Northwest forests (McKane and Tingey 1997) To evaluate the regional applicability of MBL-GEM to Pacific Northwest forests, we require sites that are geographically and ecologically distant from the sites being used to parameterize the model, i.e., several sites in Oregon including the South Santiam watershed, Cascade Head and H.J. Andrews Experimental Forest. The Olympic National Park in western Washington and the eastern Cascades of Oregon are ideal sites for model evaluation because they occur in areas that have very different climates, soils, and/or vegetation than the sites used to parameterize MBL-GEM. The range of conditions provided by these sites provide a good opportunity for developing a regionally robust biogeochemical model for the Pacific Northwest. The data from the Olympic National Park will provide an important test of the reliability of the regional model parameterization as it contains a larger range of conditions (e.g., precipitation, growing season, elevation, etc.) than for which the model was parameterized.

To accomplish the regional validation objectives we propose to carry out two primary activities under the DISPro Project:

- Collect biogeochemical and meteorological data at four relatively pristine sites in the Olympic National Park and at a site on the east side of the Oregon Cascades preliminary to submitting a proposal to work in Kings Canyon/Sequoia or Glacier National Park(s) to evaluate the regional applicability of MBL-GEM. The data and data collection methods as described in Section 3.2.1 (Task 1), will be used with this project.

- Make additional improvements to MBL-GEM to better address risk assessment issues of interest to the EPA.

In addition to addressing DISPro goals, the research activities proposed here will contribute to a number of other risk assessment activities at the EPA, including the Forest Ecosystem Indicators Program and TERA I and II. All of these projects share the goal of making spatially explicit predictions of how environmental stressors will effect the health and sustainability of forest ecosystems throughout the Pacific

Northwest. A major product of the DISPro and Forest Indicators Program will be to produce a well-validated, single parameter set of MBL-GEM that can be applied with confidence to all major forest types, soils and climatic conditions within the Pacific Northwest.

5. PROJECT MANAGEMENT AND QUALITY ASSURANCE STATEMENT

Management

This Project is an integrated effort with contributions from many different organizations and individuals from various disciplines. Major responsibilities for management of the Project within the Terrestrial Plant Ecology Branch, of the Western Ecology Division are given below:

Branch Chief

- Ensures that research addresses needs of EPA ORD Programs (EMAP, Global Change, Tropospheric Ozone); and themes of the Western Ecology Division (rhizosphere processes, extrapolation)
- Allocates funds and EPA FTEs to Projects according to ORD Program needs
- Ensures that all publications and presentations meet the requirements of the Laboratory and Agency.

Project Leader

- Represents and manages Project within Branch
- Ensures that research addresses project hypotheses and objectives.
- Peer review manuscripts, and interacts with people within and outside the Agency (e.g., International Geosphere-Biosphere Programme, Global Change and Terrestrial Ecosystems Program)
- Allocates funds according to Project needs in terms of on-site contractors, staff from EPA's Senior Environmental Employment Program (SEE), post-docs (National Research Council), undergraduate or graduate students (National Network for Environmental Management Studies, NNEMS), other contracts or agreements, and equipment and supplies
- Ensures that all publications and presentations meet the requirements of the Project
- Facilitates scientific and administrative responsibilities within Project including regular staff meetings. The meetings include seminars and discussions where participants will discuss critical issues including the relationship between the experimental and modeling research such as model assumptions and predictions.

Principal Investigators

Task Leaders

- Represent and manage Tasks within Project
- Ensure that research addresses Task objectives.
- Determine staff and funding needs (EPA staff, contractors, SEEs, NNEMS, Post-docs) and oversees them within Task

Others

- Oversee specific task measurements, statistics
- Other EPA scientists, on site contractor Work Plan Manager, NRC Postdocs

Project Scientists

- Responsible for day-to-day implementation of research plan. Recommend equipment and supplies needs, make direct measurements, process and evaluate data, maintain facilities and equipment, contribute to and/or provide publications and presentations
- EPA, contract staff, SEEs, NRC, NNEMS, other affiliations

Administrative Assistants

- Record and distribute Principal Investigator meeting notes
- Facilitate processing of purchases
- Facilitate preparation of publications and presentations
- Maintain common Procite literature database

Quality Assurance

All data collected as part of this Project must meet EPA requirements regarding Quality Assurance (QA). This involves preparation and approval of a Quality Assurance Project Plan (QAPP), Standard Operating Procedures (SOPs), and other documentation according to the QAPP, monitoring research activities to make sure the QAPP is adhered to, and reporting indicators of data quality to management Scientists in this Project have the responsibilities regarding QA given below.

Branch Chief

- Ensures that all technical outputs meet the QA requirements of the Division and Agency.

Project Leader

- Is responsible for overall QA performance and coordination of the Project. This includes primary responsibility for QAPP and approval of specific SOPs. Works with PIs to evaluate and maintain quality of data in Project. Interacts with Division QA staff in terms of official audits.

Principal Investigators

- Are responsible for carrying out entire or portions of the Project Research Tasks and insuring the quality of the results generated from the tasks. This includes contributing to the QAPP and primary responsibility for evaluating the quality of data measured under specific SOPs.

Project Scientists

- Assist in writing SOPs and have primary responsibilities for implementing SOPs as they perform measurements.

Branch Administrative Assistant

- Assist the Project Leader in managing QA documents.

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APPENDIX A

Description of soil profiles at the Intensive Sites in the western Cascades.

Falls Creek Site

The soil pit was located on a well-drained bench with a slope of about 2 - 6%. Subrounded stones within the soil profile indicate glacial transport. The weathering rinds on the stones are approximately 3 mm thick. The land surface of this soil may be as old as 800,000 years (Doug Shank, pers. comm.). Soil development is fairly well expressed with strong granular A and upper B horizons with moderate organic matter content. The texture of the soil is mostly silt loam with particles of very coarse sand and very fine gravel. Table A-1 describes the soil profile for the pit in the clear-cut.

Toad Creek Road Site

The description of the soils derived from a profile in the clear cut at the High Site is given in Table A-2.

Table A-1. Soils description for the Falls Creek Site.

Horizon designation	Depth (upper-lower) (cm)	Bulk density ($g\ cm^{-3}$)	Description (based upon a description by J. Kern, April 1993)
O1	2 - 0		Fibric material, organic layer, few fine and medium roots
A	0 - 8	0.86	Loam to coarse silt loam with some sand, weak fine and medium subangular blocky and medium to strong granular structure, very dark brown, many fine and medium roots
BA	8 - 13		Silt loam with very fine gravel, very dark grayish brown and dark brown, strong fine and medium granular structure, many medium roots
Bw1	13 - 33	0.96	Silt loam with very fine gravel, very dark grayish brown and dark brown, moderate fine and medium granular structure, common medium and few coarse roots
Bw2	33 - 55	0.96	Silt loam with very fine gravel, dark brown, weak fine and medium granular structure, very few medium roots
C	55+		Silt loam, dark brown, massive structure, very few fine roots

Table A-2 Soils description for the Toad Creek Road Site.

Horizon designation	Depth (upper-lower) (cm)	Bulk density ($g\ cm^{-3}$)	Description (based upon a descriptions by J. Kern, April 1993 and D. Lammers, summer 1994)
O_1	6 - 4		Needles and twigs
Oa	4 - 0		Decomposed needles and twigs
A1	0 - 13	0.85	Sandy loam, weak medium subangular blocky structure, dark reddish brown color, few fine to medium roots
A2	13 - 38	0.82	Sandy loam, weak medium subangular blocky structure, dark reddish brown color, many very fine roots
AB	38 - 52	0.78	Sandy loam, weak medium subangular blocky structure, dark reddish brown color, common very fine to coarse roots
Bw1	52 - 92	0.78	Gravely, sandy loam, weak medium subangular blocky structure, dark reddish brown color, few fine to coarse roots
Bw2	92 - 112		Cobbelly, sandy loam, weak medium angular blocky structure, dark reddish brown color, very few fine and medium roots
Bw3	112 - 136		Gravely, loam, weak fine single grain structure, dark brown color, few fine and medium roots
Bw4	136 - 166		Extremely gravely, loam, weak fine single grain structure, dark brown color, very few fine roots

APPENDIX B

Description of the extensive field sites

There are a number of existing research sites in the Pacific Northwest Forests operated by a number of different agencies and universities that may be suitable for out needs. Where possible, our Research Project will take advantage of established research sites. Existing sites can reduce our costs of data collection if they have the appropriate data and have the appropriate ecological characteristics.

The H.J. Andrews LTER - The Andrews LTER is located in the western Cascades in the McKenzie River Basin. Elevation in the Andrews forest ranges from 410 to 1630 m. At the main meteorological station at 430 m elevation, mean monthly temperature ranges from 1 (January) to 18 C (July). Average annual precipitation varies from 2300 mm at the base of the Andrews to 3550 mm at upper elevations. Forest types in the Andrews are typical of those described above for the intensive sites, i.e., Douglas fir, western hemlock, and western red cedar dominate at the lower elevations, with shifts to Pacific silver fir and noble fir at higher elevations. About 40% of the Andrews is in old-growth stands (dominant trees > 400 yr-old), about 20% in mature stands that originated from wildfires (100 to 140 yr-old), and the remainder in stands of various ages, composition and stocking densities resulting from clearcutting and shelterwood cutting since 1950. The main theme of ecosystem research in the Andrews is to understand and contrast the effects of land use, natural disturbances, and climate change on key ecosystem properties of hydrology, C and N cycling, biological diversity, albedo, trace gas exchange, and site productivity, topics that mesh well with some of the research proposed here.

Cascade Center for Ecosystem Management Permanent Study Plots - The Cascade Center for Ecosystem Management oversees an extensive network of over 100 permanent vegetation plots in Oregon, Washington and other western states. Ongoing measurements are being conducted on the dynamics of tree and understory species in young, mature and old-growth forests. These long-term data describe the processes of succession, tree mortality, canopy gap formation, biomass accumulation, dynamics of coarse woody debris, and timber growth. We are working with Drs. M. Harmon and S. Acker of Oregon State University at a subset of these sites to obtain geographically extensive data that can be used to develop a regionally robust parameterization of the biogeochemical model. The specific sites that we are studying include the Cascade Head Experimental Forest near the Oregon Coast, the H.J. Andrews Experimental Forest and the Middle Santiam Research Natural Area in the west Cascades, and the Metolius Research Natural Area in the east Cascades. The west-to-east transect represented by these sites captures a wide range of climatic and edaphic conditions. For example, the east end of the transect (Metolius) has a mean annual temperature 3 °C cooler than the west end (Cascade Head), receives 1/4 as much precipitation, and has substantially less total soil

nitrogen. As a result of these differences, there are significant differences in stemwood production and biomass accumulation across this transect. At Cascade Head, we will study two plots, a 150-yr-old Douglas-fir/western hemlock stand and a 150-yr-old sitka spruce stand, both of which are highly productive. At H.J. Andrews, we will use two mature Douglas-fir/western hemlock stands. The Middle Santiam RNA study plot is in an old-growth (>500 yr) stand of Douglas fir that has one of the highest standing biomasses reported for the Pacific Northwest. The Metolius RNA study plot consists of slow-growing, old-growth Ponderosa Pine. In addition to the existing long-term biomass data, total soil carbon and nitrogen stocks have recently been measured for all of these sites. We will also collect litterfall data for at least one year at the Cascade Head and Metolius sites.

Olympic National Park - The University of Washington has been conducting research on C and N cycling in the National Park for more than a decade. These plots in addition to other plots being established by EPA, Olympic National Park and USGS will provide data on C and N cycling under a range of vegetation and climate conditions. The Park offers a unique opportunity to utilize a number of gradients to test model performance and also specific hypotheses. The influence of a range of vegetation types on model performance will use data East Twin Creek (Hoh River) and the Quinault River (Sitka spruce, Douglas-fir hemlock) and Hurricane Ridge (Subapline fir). Also the model will be tested using sites differing in precipitation: High Precipitation, more than 4 meters annually (East Twin Creek (Hoh River) and the Quinault River); Low Precipitation, less than 2 meters annually (Deer Park).

Long-Term Ecosystem Productivity - Long-term research sites for the USFS LTEP are located in the Siskiyou, Willamette, and Siuslaw National Forests in Oregon and in the Wenatchee National Forest and on Washington state land in the Olympic National Forest in Washington. Many of these forests are generally of a similar type as our intensive study sites, but grow under different climatic and edaphic conditions. The major scientific questions being addressed by research in these areas is how species composition and organic matter affect a wide range of ecosystem patterns, processes, and ecosystem productivity over the long term (200 yr; B. Bormann, pers. comm). These questions will be addressed by the use of replicated stand-scale experimental manipulations which represent a range of possible forest management practices, including likely organic matter removals and establishment of different successional plant assemblages.

Small-Plot Manipulations - In addition to the stand-scale manipulations, small-plot manipulations are being installed in the Willamette LTEP research site (B. Bormann, pers. comm.). The major focus of the small-plot studies is to examine how mulched organic matter originating from a wide variety of different plant species and parts will change soil properties and growth of Douglas fir seedlings over a 5-10 yr period. These experimental small-plot sites will provide ideal locations to test hypotheses for the development of indicators related to C and N utilization by producers and consumers.

Forest Health Monitoring Program - The USFS and State Forestry departments are establishing permanent monitoring plots under the FHM program as described above (Campbell and Liegel 1996). We plan to evaluate sensitivity and variability of potential ecological tools resulting from our research in areas adjacent to the FHM plots. This will enable us to seek relationships between our process-based indicators and the FHM program's structure-based indicators.

Wind River Canopy Crane - The Wind River Canopy Crane Facility (WRCCF) is located in a research natural area (RNA) of the Gifford Pinchot National Forest, just north of the Columbia River Gorge in southern Washington. The site is cooperatively managed by the College of Forest Resources of the University of Washington, the USFS PNW Research Station, and the Gifford Pinchot National Forest Wind River Ranger District. The site is about 330 m in elevation and exemplifies the old-growth Douglas-fir and western hemlock forests (trees are estimated to be about 460 yr-old) which once covered much of the western Cascades. The climate is typical of the western Cascades with about 2500 mm precipitation per year, 2330 mm of snowfall per year, and a mean annual temperature of 8.7 C. Most precipitation falls between October and May, creating a summer drought condition. The soils are sandy loams developed in volcanic tephra over lahar and basalt bedrock. This site is unique as the crane allows access to the canopy at about 60 m above the forest floor in an area of about 2.3 ha. The site collects a full suite of micrometeorological data and has collected baseline data on forest structure.

Ponderosa Pine Research Sites - Several field sites are located in the ponderosa pine region of central and eastern Oregon that we can use for this study. These include sites in (1) Pringle Falls Experimental Forest, (2) a Research Natural Area of the Deschutes National Forest near Camp Sherman (west of Sisters), and (3) near Black Butte. The Pringle Falls Experimental Forest, south of Bend, OR, was established in 1931 as a field site to study silviculture and related topics in the conifer forests east of the Cascade Mountains, including studies on growth, insect damage, etc. The site near Camp Sherman was established to conduct a stand gas exchange experiment, the major goal being to determine the exchange of CO_2 (including separation of fluxes from understory, soil, and tree foliage) and water vapor between the atmosphere and a ponderosa pine forest using eddy correlation techniques (M. Unsworth and colleagues). This site is well instrumented for climatic variables and has a tower for canopy access. At the site near Black Butte, experiments into the response of large trees to water limitations due to environmental and hydraulic transport capacity in the ponderosa pine forest are being conducted by B Bond and M Ryan (OSU and USFS).

APPENDIX C

Application of MBL-GEM to the H. J. Andrews Forest

Data collected to date

Data that have been collected to date include a detailed characterization of C and N cycling in the low and high elevation field site forests and clearcuts (see Section 3.1.). Essentially all of the data listed in Table 3.3 (Section 3.2.1) have been measured for the Toad Creek Road and Falls Creek Cascade field sites. Most of the fluxes have been measured for more than two years and will continue to be measured for a period of at least three years. Climate data (Section 3.1) have been recorded continuously for at least a full annual cycle so that seasonal changes in carbon and nitrogen cycling can be more meaningfully interpreted. We are currently using the field data to calibrate MBL-GEM and have developed a preliminary parameterization of the model. This parameterization will be improved as additional data from these and additional sites become available.

Application of MBL-GEM: an example

As an illustration of how MBL-GEM can be used to assess short- and long-term effects of disturbance on ecosystem C and N dynamics, we describe here a simulation of the effects of harvest on an old-growth Douglas-fir ecosystem. For this example, we parameterized the model for a 450 yr-old Douglas-fir forest at the H.J. Andrews Experimental Forest in Oregon (Grier and Logan 1977, Sollins et al. 1980). This parameterization was then used to simulate the effects of three consecutive harvests spaced 60 years apart. At each harvest, 50% of living biomass was removed as forest products, 40% of the pre-harvest biomass was converted to logging slash and added to the detritus pool, and living biomass was reduced to 10% of the pre-harvest biomass. After each harvest the model was run for 60 years assuming no change in present-day climate.

The model results suggest that this harvest regime appreciably reduces ecosystem C (Fig. C-1a) and N (Fig. C-1b) stocks, a result attributable to both the removal of forest products and to increased rates of soil respiration (Fig. C-1c) and N leaching (Fig. C-1d). The recovery of vegetation C during forest regrowth steadily declines for consecutive harvests. Figure C-2 summarizes the simulated cumulative effects of the three harvests, showing that after 180 years total ecosystem C decreased by almost 60%, while decreases in ecosystem N, NPP and net N mineralization approached 20%.

These results suggest that frequent and intense harvests may have a substantial impact on ecosystem productivity and sustainability, however, much work remains to be done to constrain and test the model. Our purpose here is to illustrate the use of a process-based model for synthesizing information on ecosystem processes and on how those processes interact over time. It is because of this synthesis that process-based models surpass any other method of projecting responses to environmental change.

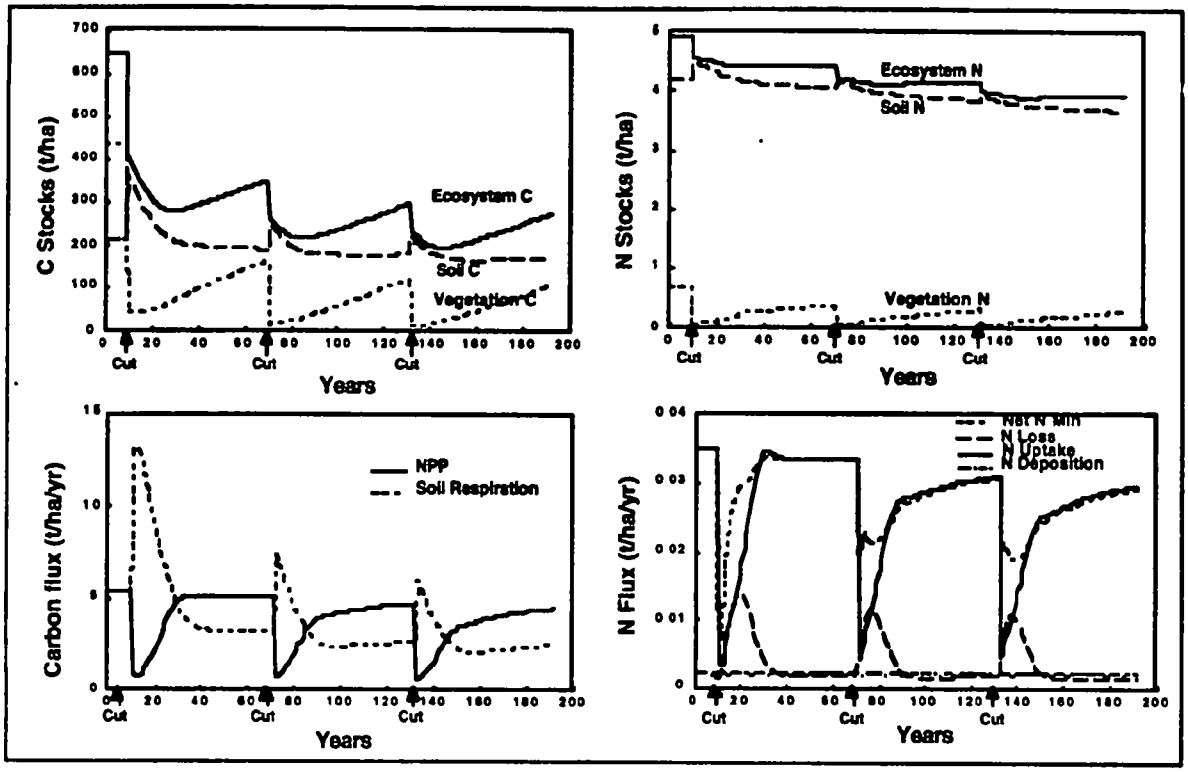

Figure C-1. MBL-GEM simulation of 3 consesecutive clear cuts. Parameterized for the H.J. Andrews LTER Site.

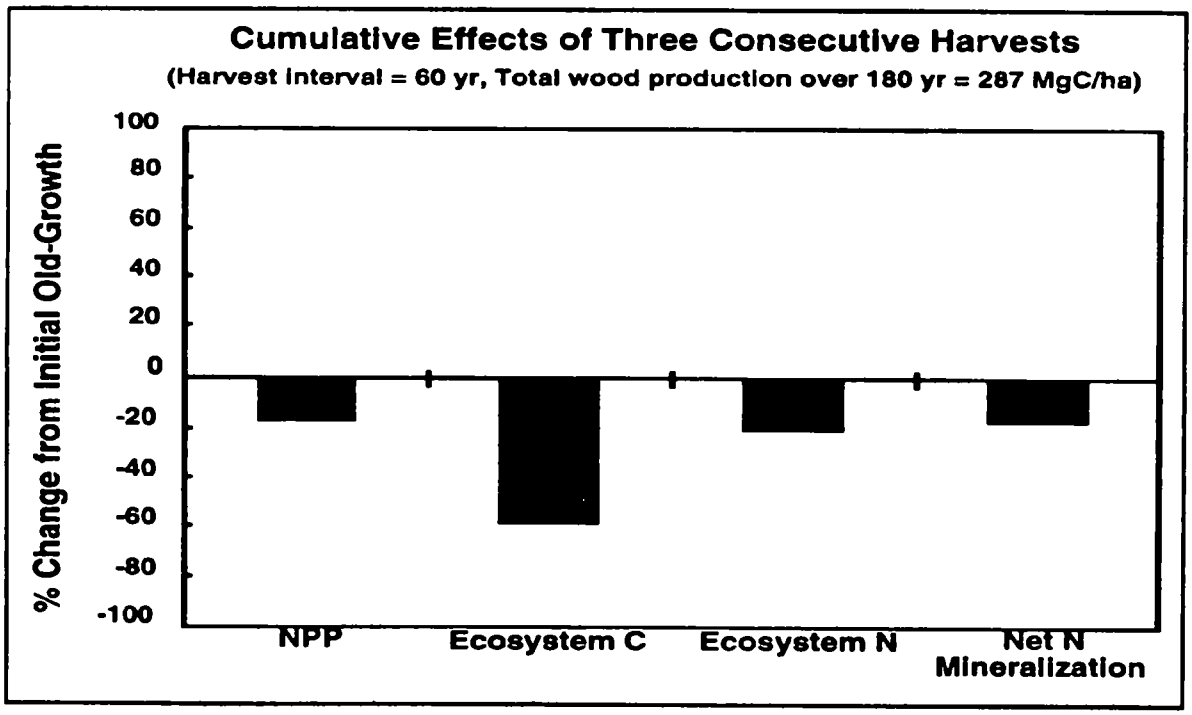

Figure C-2. Summarizes the simulated cumulative effects of the three harvests, showing that after 180 years total ecosystem C.