CORE
Metadata, citation and similar papers at core.ac.uk
Provided by The University of Utah: J. Willard Marriott Digital Library

2 The study of ecosystems in the context of global change

E.-D. Schulze, R.J. Scholes, J.R. Ehleringer, L.A. Hunt, J. Canadell, F.S. Chapin III and W.L. Steffen

2.1 Introduction

Global change research requires not only knowledge of how individual species (e.g. pests) respond to climate and land-use change, but also an understanding of the responses of whole systems to their multiple and interacting drivers. An upscaling from single systems to landscapes and continents is an additional essential component of global change research. In contrast to the well-established research methodology in the ecophysiology of species (see Pearcy *et al.*, 1989), the approaches to studying ecosystems as a whole, and the theory required to identify key parameters that drive the multiple interactions at the ecosystem level, are less developed (see Schulze, 1995*a*). The available tools become even more limited for the study of responses at geographically broader scales. One of IGBP's (International Geosphere – Biosphere Programme) products has been the contribution to methodology and theory for extrapolations from ecosystems to continental scales.

Experimental investigations of ecosystems started between 1960 and 1970 with the International Biological Programme (IBP), and the ecosystem-level global change studies of GCTE (Global Change and Terrestrial Ecosystems) have built strongly on that heritage. Global change research has reiterated that ecosystems are difficult to study for several reasons:

Large-scale processes: Important ecosystem processes, such as biogeochemical cycles, disturbance regimes and demographics, take place at scales much larger than the typical experimental plot of a few hectares. These large-scale processes are only partially manifested at the smaller scale.

Slow and often highly buffered processes: Many ecological processes, including succession and changes in the soil, require decades to centuries for completion and may even be in a transitional dynamic state. Nevertheless, there may be no perceptible change for many years following the application of a treatment, but rapid change may occur once the buffering

capacity of the system is exhausted. Ecosystem changes may not be fully revealed even in the lifetime of the researcher, which is a very different scale than the average time of support for typical research projects.

Complexity: Ecosystems contain unknown numbers of organisms. This results in large numbers of variables that affect ecosystem fluxes, and these variables are connected to one another typically in nonlinear relationships. This makes it difficult to isolate single factors to be systematically varied in an experiment without simultaneously disrupting many others. The number of interactions swiftly renders factorial experiments unwieldly, especially since the nonlinearity of the responses requires several treatment levels for each factor, rather than just a 'high or low' or 'present or absent' treatment.

Fluxes are difficult to measure: Ecosystem processes often cannot be directly observed but must be inferred from their consequences. For material flows, this usually involves observing changes over time in pools. Where the pool is large relative to the flow, the small result is easily lost in the noise created by sampling and analytical variation. In addition, the net flux in and out of systems does not reveal the internal circulation of substances.

True replication is difficult: Classical statistics assume multiple identical units, randomly assigned to treatments. The high spatial heterogeneity characteristic of ecosystems means that every patch is in some respect unique. Furthermore, the experimental treatment is often applied by nature, rather than by the researcher, and is thus not randomly assigned. Finally, there are ethical constraints in applying large-scale manipulative experiments to what are frequently already-threatened ecosystems. Truly global experiments (such as the inadvertent experiment we know as 'global change') have no control and no replicates, but contain only a time series in a naturally highly variable environment. Conducting such an experiment may threaten not only our own well-being, but also that of millions of other species.

Based on these intrinsic problems, ecosystem research started in the IBP, including some of the most important long-term observations of ecosystem pools and processes, is still used by IGBP. Examples are the long-term observations of soil solution chemistry and acidification (Ulrich, 1989) or the monitoring of the atmospheric composition of CO_2 (Conway *et al.*, 1994). While IBP was focused on understanding mainly productivity, the study of ecosystems emerged in the following decade of acid rain and air pollution research. In this period it was demonstrated for the first time that phenomena such as forest decline cannot be explained by single processes and linear regressions. Rather, to interpret the decline in forest health adequately, a systems approach was

needed that included direct and indirect effects, feedbacks and interactions (Last & Watling, 1991). Air pollution research demonstrated also the necessity of ecosystem-scale experiments in addition to the field observations (Mooney *et al.*, 1991). Thus, the following earlier approaches are now used for the study of ecosystems in the context of global change: (i) investigations of natural extreme events (e.g. volcanic eruptions, El Niño); (ii) long-term observations at the plot level; and (iii) ecosystem manipulation (Mooney *et al.*, 1991). The global scale of the questions addressed by IGBP required an additional tool. It is impossible to study all ecosystems globally with the same level of detail. This led to the idea of establishing continental-scale transects as a basis for replicated observations (see Chapter 4).

This chapter gives an overview of the research approaches and techniques used and developed by GCTE, from the plot-level to the global scale. Databases and model development will be discussed in Chapters 5 and 6, respectively.

2.2 Approaches to the study of ecosystems

Figure 2.1 presents a schematic summary of approaches to the study of ecosystems in the context of global change. In the following sections each type of approach is described with a number of examples of its use and a discussion of its strengths and weaknesses.

2.2.1 Sampling problems in global change research

Every study plot is a unique sample from a highly variable environment. For example, when a forest transect through Europe was studied (NIPHYS/CANIF project – GCTE, 1996), plots were selected so as to maintain soil conditions as similar as possible in order to detect effects of climate and nitrogen deposition. Following the selection, it emerged that the local variability in nitrogen concentrations of needles and leaves was larger than the variability along the transect as a whole (Bauer *et al.*, 1997). The same problem occurs again, although at a different scale, with respect to the heterogeneity within a plot.

Oren *et al.* (1989) made use of this variability at the plot level in order to extract general regression relationships that describe the variation in a functional manner. It became obvious that a series of smaller plots, representing local variability, is more useful than the study of a single large plot. Another approach using regional variability for a mechanistic understanding of processes was that of Durka *et al.* (1994). Based on the regional variability of 360 forest surface springs, six plots were identified that represented the regional range of conditions, and for these plots those processes that caused the observed

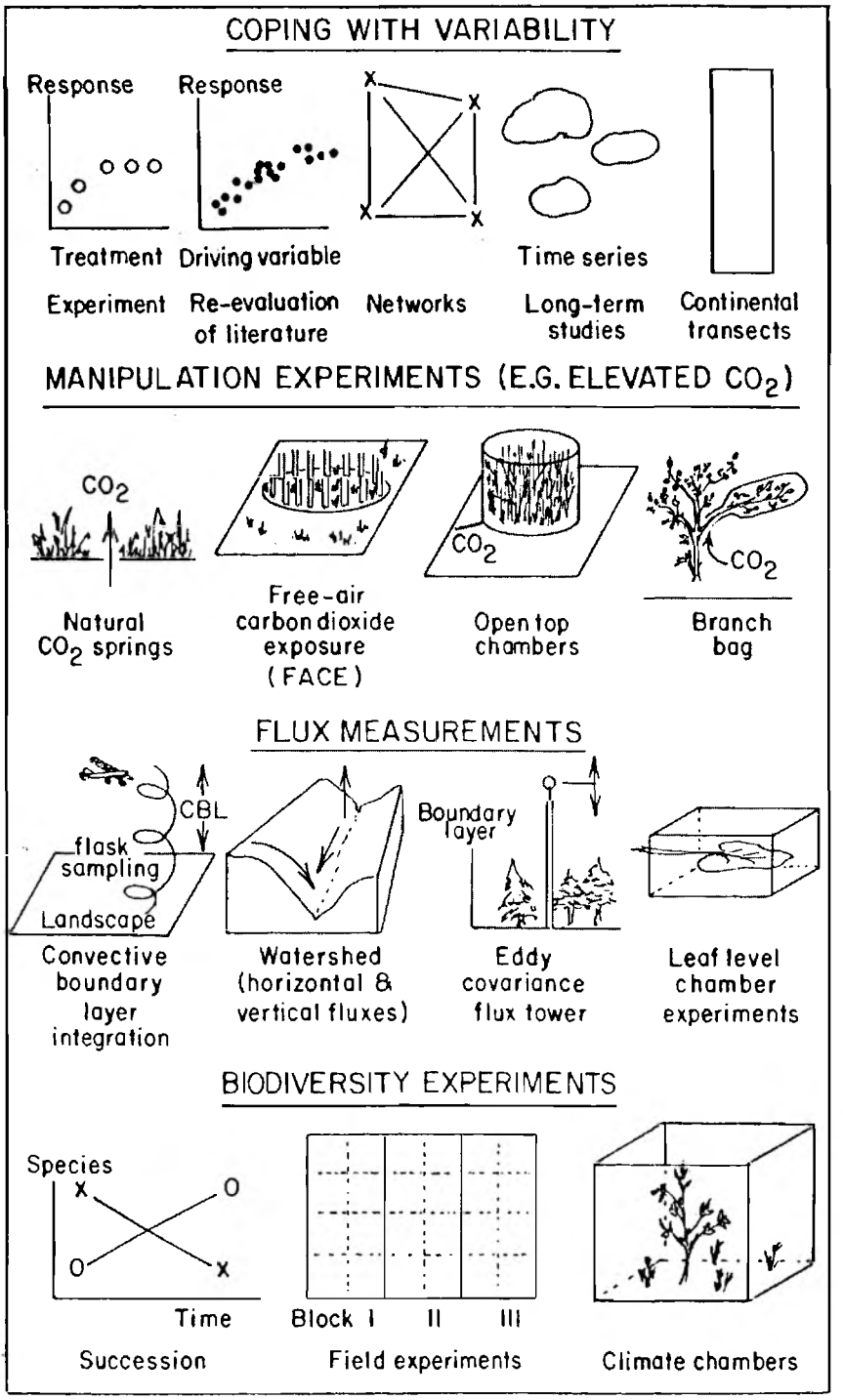

Figure 2.1 A summary of approaches to the study of ecosystems in the context of global change. Each of the approaches is described in more detail in the text.

variation in outputs were identified. Thus, local versus regional or continental variability can help in identifying key processes. However, there are limits to this approach. Hornung (1992) studied some 40 watersheds throughout Europe, but it became difficult to identify single processes that explained the variability, because the number of parameters causing change in a network of sites (not a transect) was greater than the number of plots being studied.

The statistical problems of how to deal with variability at different scales are a serious issue for global change research, and have not been resolved. The Long-Term Ecological Research Program (LTER) of the United States, the Chinese Ecological Research Network (CERN) and the biosphere reserves of Russia are networks of sites representative of biomes. They can serve as ground truth for global modelling, but a comparison of these sites cannot produce response curves for variables that drive the change from one biome type into another because their selection was biome- and not gradient-oriented. This was one of the reasons that IGBP adopted the transect approach which allows an *a priori* definition of a driving variable against which the responses (often nonlinear) of ecosystem parameters can be examined (see Chapter 4).

In view of the immense problem of local and regional heterogeneity, an approach has emerged in GCTE for ecosystem studies at global scale. Processes have been studied, independently of plots, by focusing on the ecology of species. For example, the re-examination of data on CO_2 and water exchange of plants and their assessment in relation to other driving variables, such as nutrition, has led to new information about the general relations between parameters that can then be tested at a plot or transect scale (Körner, 1994; Woodward & Smith, 1994*a*; Schulze, 1994*c*). This approach has the advantage that it contains, by definition, species- and habitat-related variability. This gives more power for the use of these functions in global models than functions based on observations on a single plot. The disadvantage of this approach is also obvious; the variables are not measured at the same time on the same object. It is necessary to integrate this information, for example, according to plant functional types (see Section 2.2.2 and Chapter 8). The combination of (i) development of general and global relations; (ii) their test in specific situations; and (iii) their use in modelling on landscape or regional scales has become a new approach in the context of global change research.

With respect to ecosystem experiments, a different set of statistical constraints and considerations emerge (Jeffers, 1993). Classical statistics, such as the Analysis of Variance (ANOVA), were developed for the analysis of agricultural trials, where true replications, factor separation and discrete treatment levels are possible. This design proved to be successful in applied research. In fact, a treatment effect can readily be produced if the treatment is strong enough in relation to the control. For many ecological applications where these condi-

tions are not met, the more general approach offered by regression analysis (including nonlinear and multivariate forms) is more appropriate. In general, the focus in ecological studies is less on proving that two treatments are statistically different from one another, but on establishing the form of the relationship between dependent and independent variables, and identifying critical thresholds. Where the nature of the treatment is not known *a priori* – as is frequently the case in gradient studies – there are several indirect gradient techniques to reveal the relationship between environmental drivers and ecosystem responses. Thus, for ecosystem experiments in the context of global change, it is more appropriate to quantify a response surface than a particular treatment effect.

Numerical simulation modelling is indispensable for exploring the interactions in even moderately complex systems, especially where nonlinearities and feedbacks render them intractable to analytical mathematical techniques. Furthermore, models act as hypotheses which test whether current understanding of the processes at work is sufficient to recreate the observed phenomena. This set of techniques has become so important for global change research on terrestrial ecosystems that a full chapter – Chapter 6 – is devoted to a treatment of the topic.

2.2.2 Ecosystem comparative studies

Biome comparisons and networks

The comparison of widely separated ecosystems which are acknowledged to be different in their details provides a test of the robustness of our understanding of systems in relation to soils and climate. The approach is especially powerful when one treatment is repeated in several sites, across a range of environments and ecosystems. This obviously requires standardization of treatments and measurements. An example is the experiment designed by the GCTE Rice Network for implementation at four tropical and three temperate sites (Ingram, 1995). The experiment involves determining the effects of higher temperatures and nitrogen application levels on the grain yield of rice. The association of related sites into networks has become an important component of international collaborative science efforts (see also Chapter 3).

The difficulty of replicating an ecosystem experiment spatially may sometimes be avoided by replicating in time instead if all other conditions are constant. The main problem with this approach is in ensuring that the time-separated treatments are independent of each other, and that factors such as climate differences are accounted for. The comparison of observations taken over long periods of time within one ecosystem is a powerful technique for detecting change, provided that the statistical techniques appropriate to time

series analysis are used and that methodological consistency in observation techniques is maintained. The time series of soil solution chemistry in the Solling study of Ulrich is a classical example of this approach (Ulrich, 1989). The LTER sites in the USA and the CERN sites in China have been set up for this purpose.

Very long-term processes can be explored using paleoecological data. For example, the rate of species migrations has largely been inferred from palynological records of the changing distribution of species in response to past climate (Firbas, 1949). In an analogous way, the growth rate of trees over the past few centuries can be inferred from tree rings (e.g. Spieker *et al.*, 1996).

Functional types

It is practically impossible to study all species involved in an ecological process on an individual basis. One approach to tackling this problem is to classify species according to their functions rather than using the traditional phylogenetic approach. While the study of functional types goes back to the beginning of ecology in the last century (see Schulze, 1982 for a review), GCTE has taken up the challenge to develop a scheme of functional types for process-related studies (Woodward & Cramer, 1996*b*; Smith *et al.*, 1997). It emerges that structural characteristics have an overriding importance for classifications based on physiology. In fact, the classification by Monsi (1960) may encompass most requirements (annual/perennial; herbaceous/woody; evergreen/deciduous), except for a few physiological traits such as C_4/C_3 metabolism and nitrogen fixation, which add a new physiological quality.

Although the use of functional types is proving useful in global modelling (see Chapter 8), studies of community ecology have demonstrated that individual species also matter in a global change context (Mooney *et al.*, 1995). A good example is rooting depth. In grasslands, 90% of the root biomass occurs in the top 20 cm of the soil profile (Jackson *et al.*, 1996*b*), while the average maximum rooting depth reaches 3 m (Canadell *et al.*, 1996*a*). From a closer inspection of the grassland data from the root atlas by Kutschera (1960), it emerges that 187 species reached 70 cm rooting depth on average, but 10 of the 187 species reached 223 cm. In case of increasing drought, those 10 species have a much larger chance of survival because of this species-related trait.

Certainly a new functional type could be defined for deep-rooted herbaceous species. However, if functional groups are defined for all parameters that are important at the community level (e.g. fecundity, leaf orientation, phenology, etc.) a species-level classification may result, because the many different combinations of the functional attributes will eventually describe a specific species. Thus, the earlier expectation that species-specific studies could be avoided is dampened with the knowledge that combinations of species-specific traits are

often important in understanding the response of terrestrial ecosystems to global change (see also Chapter 11). The problem can be resolved to some extent by defining two kinds of functional types – 'control' and 'response' types (Walker, 1997). The importance of species has been specifically a problem with pests and parasites, which almost always act by a species–species interaction. Nevertheless, in many cases for testing focused hypothesis at the ecosystem level, functional types remain an important research tool.

Gradient studies

Observing ecological responses along gradients of environmental drivers has been one of the basic tools of ecology since the earliest years of the discipline. Many ecological responses in nature take the form of a continuum, rather than discrete steps, making a gradient a natural way to examine them. Gradient analysis is also a way of using spatial variability for testing hypotheses. A long-established natural gradient allows for the long adaptation times necessary for many important processes to adjust to the gradient, which would not be possible to recreate in a newly established treatment. In many cases the gradients are complex (involving more than one driving variable, varying either in parallel or independently). Sometimes the gradients are continuous in space, as, for example, along the catena sequence of soils that form down a slope (Giesler *et al.*, 1998). However, more often gradients are not continuous in space. A special case of a gradient study is the chronological sequence of sites of different age since disturbance (e.g. Schulze *et al.*, 1995).

Carbon dioxide emitted by natural springs diffuses away from the source, providing a gradient of elevated atmospheric CO_2 which has proved unique for examining the long-term effect of elevated CO_2 on vegetation (Koch, 1993; Miglietta *et al.*, 1993). Variations in wind speed and direction disrupt the diffusing plume, causing large fluctuations of the CO_2 concentration at any given point, but averaged over the day or the growing period of a plant, there are clear patterns of exposure. Gradients of partial pressure of CO_2 can also be obtained with increasing altitude in mountains, but in this case the gradient is confounded with increasing UV-B and decreasing temperatures, and often with changes in precipitation. Altitudinal gradient studies play a key role in a major IGBP research effort on global change in mountainous regions (Becker & Bugmann, 1997).

GCTE adapted the well-established ecological technique of gradient analysis to the global scale by developing the idea of 'megatransects', a concept subsequently adopted by the IGBP as an inter-core project research platform (Koch *et al.*, 1995*a*,*b*). The rationale for the IGBP transects and their use in global change research is described more fully in Chapter 4. An example of the use of

the transects for analysing the interactive effects of global change on terrestrial ecosystems is presented in Chapter 12.

2.3 Manipulative experiments

Building on knowledge from forestry and agriculture, ecosystem-level manipulative experiments were started in the period of IBP (Tamm, 1991), where one major question was a global quantification of productivity. However, the reservations of ecologists about such experiments have often been cited: (i) There may be indirect treatment effects which make it impossible to manipulate just a single factor. For instance, the effect of fertilization on yield may be overlaid by its effects on humus decomposition, soil acidification and pollution of ground water, which, at the ecosystem level, override the immediate effect of fertilizer on growth. (ii) The history of processes on a plot may influence the result (e.g. fire history; a dead animal may have been decomposing by the tree selected for study). Results may thus be idiosyncratic and of a correlative nature rather than reflecting mechanisms.

Nevertheless, manipulative experiments at the ecosystem level are becoming an increasingly important tool in global change research. This section describes such experiments in five key research areas – effects of elevated atmospheric CO_2, impacts of warming, nutrient and water limitations, effects of ultraviolet radiation, and biodiversity interactions with global change.

2.3.1 Elevated CO_2

The fact that atmospheric carbon dioxide will continue to rise for the foreseeable future is one of the certainties in global change. Thus, a large number of studies have used experimental approaches ranging from cellular to ecosystem-level in order to predict plant and ecosystem responses to elevated CO_2. Almost all studies have focused on the effects of an instantaneous doubling of CO_2. The bias and the problems using this approach are quite obvious. A review of the published literature shows the uneven distribution of information along a response surface (McGuire *et al.*, 1995*a*). It emerges from the large body of elevated CO_2 research (see Chapter 7 and Section 12.2.2) that the hypothesis of Gifford (1994*a*) still holds, namely, that an effect at the protein level will continually shrink when investigating the same response at the leaf level, the whole plant or the ecosystem level. Thus, the approaches for studying elevated CO_2 in the context of global change must emphasize natural (low nutritional) ecosystems, with the aim of understanding the response surface of CO_2, nutrient, and water interactions at the system level.

With respect to crop species, additional problems emerge. Much recent CO_2 work has been undertaken using major crop species as the experimental target, and in particular, wheat. All such experimentation has been organized in accordance with the basic principles of research design, with randomization and replication of experimental treatments, and has endeavoured to accomodate the widely recognized impact of different base environments by continuing over at least two growing seasons. Such work has demonstrated effects of CO_2, but has not provided response curves because only high and low levels were tested in a block design, which is inappropriate for generalizations. Because of limited funds, the studies have also been conducted with a limited array of cultivars, and generally with only one cultivar in a particular study. The experiments thus do not allow for examination of genotype–environment interactions, which are generally quite significant. To help overcome this limitation, individual experiments at specific sites are being complemented by coordinated experiments in which a common genotype with seed from a standard source is examined at different sites (e.g. ESPACE, the European Stress Physiology and Climate Experiment, H.J. Jüger, personal communication). Such work is currently concentrated on one genotype; it will need to be extended to an array of genotypes if the full significance of differential genotype response to CO_2 and other environmental variables is to be fully understood.

In the absence of coordinated multi-site and multi-genotype experiments, some indication of the magnitude of the interaction terms can sometimes be obtained by analysing jointly the results from different experiments. Unfortunately, this task is often made difficult by the different modes in which data have been recorded and subsequently collated by individual researchers and research groups. To help overcome the problem, attempts have been made recently to develop some standard file structures that can be used for storing experimental details and data, for input of data into various analysis programs and simulation models, and in particular, as transfer files for the interchange of data between experimental groups (see Chapter 3 for more details).

Beyond the problem of genotype–environment interactions and of data storage and use, interpretation of the results of field experiments has often been limited by inadequate information on parts of the total system. In an attempt to overcome the problem, efforts have been made by a number of individuals concerned with agricultural experimentation to define a 'Minimum Data Set' that should be recorded in all experimental work (see Hunt *et al.*, 1994). In such a minimum data set, which perhaps could better be termed a 'balanced data set', an effort is made to encompass information on the soil, on conditions at the start of the experiment or study (e.g. soil surface conditions, soil water contents, soil inorganic N contents), on management interventions, and on various aspects of the growth and development of the plant cover. The concept is applicable to all

ecosystem studies, whether managed or natural, and should be followed widely to be able to interpret results at an ecosystem level.

The need to investigate ecosystem-level responses to elevated CO_2 has required the development of new experimental technologies (Schulze & Mooney, 1993). Most early research on effects of elevated CO_2 utilized plants grown in pots in greenhouses or growth chambers and involved exposing plants to elevated CO_2 for periods ranging from minutes up to a single growing season (Strain & Cure, 1994). While these early experiments provided important insights into short-term responses of individual plants, they revealed little about responses of entire ecosystems over long time periods. Technologies that have been developed or employed over the past decade to study ecosystem responses include ecocosms, closed chambers (CC), open-top chambers (OTC), and free-air carbon dioxide enrichment (FACE). Natural CO_2 springs can also provide a valuable understanding of long-term and evolutionary responses to elevated CO_2. Here the technologies and their respective advantages and disadvantages are briefly described in relation to sample/plot size, expense, degree of environmental control, and 'naturalness' of the system.

Ecocosms are essentially elaborate growth chambers in which investigators attempt to recreate conditions found in natural ecosystems (e.g. Körner & Arnone, 1992). This can include attempts to establish natural soil conditions and a representative assemblage of species, including microbes. A key advantage of ecocosms is that the investigator has a high degree of control over all aspects of the experiment including species composition, soil conditions, and environmental variables. The major disadvantages include the constraints on plot size and maximum plant size; the expense of constructing numerous large chambers, controlling all important environmental parameters, and operating the experiment for long time periods; and the difficulties in creating truly natural conditions.

The first attempt to evaluate *in situ* ecosystem responses to elevated CO_2 employed closed-top chambers (Oechel *et al.*, 1994). The first CCs were small and were used only in low stature ecosystems such as tundra or grassland. More recently larger chambers have been constructed for use in chaparral or even with trees. CCs are expensive to construct because of the need to control all environmental variables; operating costs may be relatively low for CO_2 but are high for the electricity needed to control the chamber environment. Chamber effects in CCs inevitably result in alteration of the radiation and wind regimes and pollinator access.

Open-top chambers were first developed for use in air pollution studies (Heagle *et al.*, 1974) and more recently have been utilized in CO_2 research. OTCs can be up to several meters in diameter and height, and thus can be used with plants as large as tree saplings. They typically are constructed of inexpen-

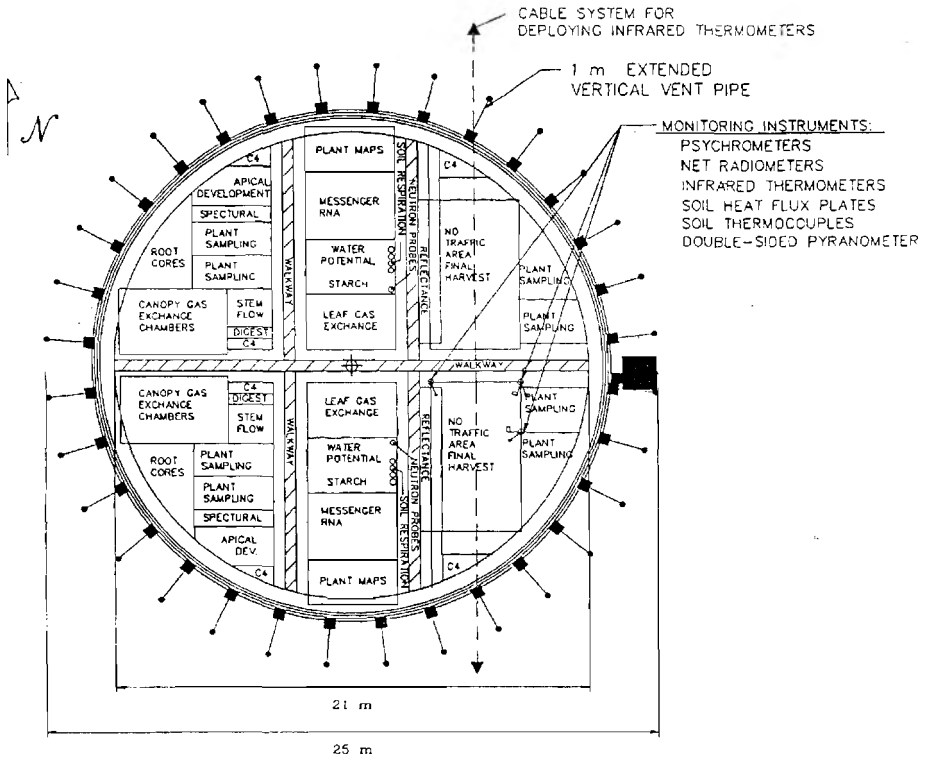

Figure 2.2 Overview of a FACE ring for the 1992–93 FACE wheat experiment in Maricopa, Arizona. Location of the assigned biological activity areas in both irrigation subplots of the ring. (From Wall & Kimball, 1993.)

sive materials (e.g. metal tubing and polyethylene sheeting) making it possible to use large numbers in factorial experiments. The concentration of CO_2 (or other gases) in the chambers is increased by adding controlled amounts of the gas to ambient air, which then is introduced to each chamber through a plenum around its base. Other environmental variables generally are not actively controlled, and natural precipitation can enter through the open top. Because of their low cost and relatively large sizes, OTCs have been used in many elevated CO_2 experiments (see Chapter 7). As with CCs, wind regimes, radiation, and pollinator access are altered within the chamber. One variant on the OTC is a tunnel system in which gradients of CO_2 are maintained, allowing plants to be exposed to a full range of CO_2 levels (Mayeux *et al.*, 1993; Hadley *et al.*, 1995). The gradient is established by blowing CO_2-enriched air through the tunnel with dilution caused by plant uptake and leakage.

The newest and best technology for investigating the responses of natural ecosystems to elevated CO_2 is FACE (Lewin *et al.*, 1992) (Fig. 2.2). In FACE

systems, CO_2 concentration within the experimental plots is elevated by introducing gas to the plots from a series of vertical pipes arranged around each plot. Wind direction, wind speed, and CO_2 sensors within the plots are used to control which pipes emit CO_2. There are two key advantages of FACE technology. First, plot size can be large (e.g. 30 m in diameter) so that a reasonable number of large plants, including relatively mature trees, can be studied in natural ecosystems. Second, since no chamber or barrier is constructed around the experimental plots, virtually all environmental factors other than CO_2 concentration are unaffected. However, during still nights, air blowers are needed to ensure a proper distribution of CO_2 over the plot, and this has been reported to have an effect on air temperature (called the 'blower effect'). The crucial disadvantages of FACE are the cost of the experimental facilities and the very high, ongoing cost of CO_2. As a consequence FACE experiments initiated to date involve few treatments and little replication. In any case, FACE is the best technology available for the most realistic field experiments calling for enrichment of intact ecosystem units. A variation on FACE is to construct a screen around the plots to reduce wind speed through the plot and thus reduce the amount of CO_2 used.

The use of natural CO_2 springs is also an important approach to study system responses to elevated CO_2. Their clear advantage is that plants native to these sites should have adapted to these conditions over long periods of time. Special caution, however, needs to be paid to the possibility that these natural springs may carry other gasses in addition to CO_2 which, even at low concentrations, may have an important effect over long periods of time. Highly variable CO_2 concentrations over time may be also a problem (Koch, 1993).

2.3.2 Warming

Over the last few years a range of techniques has been used to study the impact of global warming on ecosystem functioning. The most common techniques are buried heating cables, air-heated open-top and closed chambers, infrared heaters, and simple passive greenhouses (Fig. 2.3). A brief description of each technique is given below.

Buried heating cables have been used to increase soil temperature between 5 °C and 10 °C above ambient in various ecosystems (van Cleve *et al.*, 1990; Peterjohn *et al.*, 1993; Lükewille & Wright, 1997). This technique is commonly used in forests because the warming of an entire system would be costly and technically unfeasible in most of the cases. Because ecosystem responses to warming are expected to be largely modulated by altered soil processes, this technique can provide significant insights to better understanding of warming responses. However, the extent of the decoupling effect between the belowground (temperature treated) and the aboveground components (non-tempera-

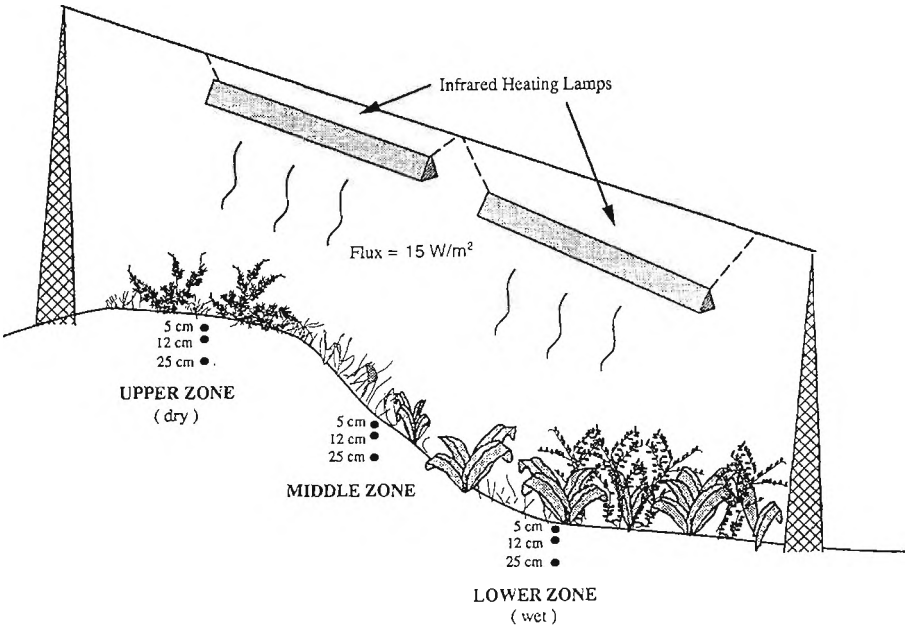

Figure 2.3 Typical heated-plot experimental profile of a montane meadow, Colorado, USA (Harte *et al.*, 1995).

ture treated) remains as one of the major concerns in this type of experiment (NSF, 1992).

Passive greenhouses have been extensively used to increase air and soil temperature (Shaver *et al.*, 1986; Debevec & MacLean, 1993; Kennedy, 1994; Wookey *et al.*, 1995; Chapin & Shaver, 1996). It is an inexpensive technique which allows extensive replication and multi-site experiments (e.g. ITEX – Welker *et al.*, 1995). However, passive greenhouses alter the micro-environment inside the enclosure, an effect which needs to be carefully monitored (Kennedy, 1995). Another problem associated with this technique is the restricted access for pollinators, dispersal agents, grazers, and pathogens.

Air-heated open-top and closed chambers have been recently used in a variety of experiments (J. Melillo & C. Neill, personal communication; S. Linder, personal communication). They provide very realistic warming, but they also suffer from the problems associated with enclosures. A variant of the open-top chamber is the gradient temperature tunnel (Hadley *et al.*, 1995). A large version of a closed chamber is the catchment enclosure used in the CLIMEX project, which studies ecosystem-level processes including soil and catchment hydrology (Jenkins & Wright, 1995; Lükewille & Wright, 1997). Because of the costly setup required for this kind of experiment, replication is usually insufficient or is lacking.

Infrared heaters suspended over relatively short vegetation are also a very

useful technique (Harte *et al.*, 1995). Infrared radiation warms the surface of plants and soils directly but does not warm the surrounding air, which has some drawbacks for reproducing the most realistic warming of a convective nature. This technique, however, allows the study of soil processes in addition to plant community dynamics (Harte & Shaw, 1995).

Another technique that has been successfully adopted is the use of a pipe grid with inside running water that controls the temperature of the soil and the immediate air above the soil surface (Hillier *et al.*, 1994).

2.3.3 Nutrient and water interactions

The potentially large impact of global change on the hydrological and biogeochemical cycles has prompted many manipulative experiments examining the effect of irrigation and fertilization on ecosystems. Such experiments have long been used to design management regimes for achieving optimal crop yield, and have more recently been applied to non-agricultural systems, including forests (Tamm, 1991). These experiments normally have a factorial design with treatments consisting of irrigation only, fertilization only, and both irrigation and fertilization, along with control plots. Both treatments are usually applied at several levels to determine the effect on the system response, with fertilizer applied either in liquid or solid forms. An innovative approach to study the interactive effects of CO_2, temperature, nutrients and water on forests is based on the addition of a soil-warming experiment and whole tree chambers with CO_2 and temperature control to irrigation and fertilization studies on Norway spruce at Flakaliden, Sweden (Linder, 1987, 1995).

2.3.4 UV-B

Concern over the effect of increasing UV-B radiation (the biologically most harmful component of the ultraviolet band) resulting from decreasing stratospheric ozone concentrations has led to numerous experiments studying the effects of enhanced UV-B on terrestrial ecosystems. Such experiments are often carried out in growth chambers or greenhouses, but more recently there has been an increasing number of field studies in more natural conditions (Caldwell *et al.*, 1995). Growth chambers have the advantage that other factors, such as temperature and humidity, can be controlled or manipulated. However, both UV-A and visible radiation are usually less than in sunlight.

UV-B radiation is controlled either by increasing it over ambient by use of fluorescent lamps or by filtering natural light to remove UV-B using either plastic filters or ozone gas. When natural light is filtered, the comparison is made between systems being exposed to ambient levels of UV-B and those exposed to reduced levels (Barnes *et al.*, 1995; Flint & Caldwell, 1996). Experiments are now being undertaken to examine the interactive effects of enhanced

UV-B with other global change factors. For example, a joint British–Swedish experiment is using lamps over OTCs to study the interactive effects of elevated CO_2 and UV-B on mountain heath vegetation (Caldwell *et al.*, 1995).

In any experiment it is important to achieve a realistic balance between UV-B (280–315 nm), UV-A (315–400 nm) and visible (400–700 nm) radiation. Visible radiation can have strong ameliorative effects and so experiments undertaken in chambers, which reduce the amount of ambient sunlight, need to ensure that supplement radiation in the 315–700 nm range is supplied. Another important consideration, especially in field experiments, is to adjust radiation for cloudiness and other atmospheric conditions; otherwise exaggerated UV-B effects may be found (Caldwell *et al.*, 1995).

2.3.5 Biodiversity

The growing recognition that ecosystems are not simply uniform surfaces acting as sources and sinks of fluxes (the 'green slime' concept), but that species diversity within ecosystems is a major factor influencing ecosystem functioning (see Chapter 11), has led to a novel type of global change experiment. Based on the earlier Rothamsted (UK) grassland experiment, in which various treatments were maintained over more than a century and the effects on diversity and species composition observed, Tilman (1993) initiated experiments in which species were exposed to a new competitive environment of neighbours. Naeem *et al.* (1994) extended this design to a statistical random selection of species from a suite within a natural community. The selected species were then grown in various combinations of density and species number with the aim of quantifying the effects of species numbers on ecosystem fluxes, a novel connection between ecosystem complexity and material fluxes (ecosystem functioning). Initial results show effects of species composition on drought tolerance (Tilman & Downing, 1994) and on ecosystem processes such as nitrate leaching (Tilman *et al.*, 1996). The problem of co-varying confounding factors in the experiments has, however, drawn criticisms of the interpretation of the results. For example, Huston (1997) re-evaluated some of the data and showed that it is not the biodiversity *per se* that drives the observed outputs, but rather stand biomass, density and other structural components of the vegetation which are correlated with the biodiversity treatment. In order to avoid such indirect responses, the recent BIODEPTH (Biodiversity and Ecological Processes in Terrestrial Herbaceous Ecosystems) experiment (J.H. Lawton, personal communication) is based on a statistical and repeated selection of species groups from a natural community. A more detailed account of the current understanding of the relationship between ecological complexity and ecosystem functioning is given in Chapter 11.

2.4 Observational studies

Despite the major advances and innovations in ecosystem-level experimental techniques described above, there are nevertheless significant remaining methodological difficulties and considerable expense associated with manipulative experiments. Thus, observational studies of ecosystems continue to play a crucial role in global change research.

Observational studies are a central activity in many areas of global change research, in GCTE and also in atmospheric chemistry (rates of trace gas emissions), hydrology (rates of evapotranspiration from ecosystems) and coastal zone studies (delivery of sediments and nutrients from upland areas). Technologies for observing ecosystem functioning, such as eddy correlation, aircraft transects and satellite observations for measuring the interaction between terrestrial ecosystems and the atmosphere, are advancing rapidly and have been described elsewhere (e.g. Chen & Cihlar, 1996; Hall *et al.*, 1996). This section emphasizes the interactions within the ecosystem itself, those that must be taken into account in designing and interpreting observational studies of ecosystem functioning, especially its interaction with the abiotic environment.

2.4.1 Pools and fluxes

Quantifying changes in the pools and fluxes of important elements is fundamental to understanding global change effects on the functioning of terrestrial ecosystems. Here some common problems associated with measuring ecosystem pools and fluxes are described, following which several recent innovations designed to overcome some of these problems are briefly presented.

Problems with a non-systems approach (components only)

At a global scale, the estimates of the size and location of the important biogeochemical pools in terrestrial ecosystems, and of their rates of change, are still inadequate, hence the continuing conjecture about the 'missing sink' (see Chapters 7, 10 and 12). Early global change studies emphasized only carbon and not the associated fluxes and pools of nutrients or water. Very often only parts of ecosystems were investigated (e.g. soil carbon, neglecting the humus layer and aboveground biomass). Other parameters important for ecosystem fluxes are still under debate (e.g. what is the Leaf Area Index of a *Sphagnum* bog?). Thus, any ecosystem study will have to start with an inventory of pools and fluxes that should be as complete as possible, i.e. it should reach beyond the focused objectives of a particular study in order to create a minimum dataset which is useful in a broader global change context (see also Section 2.3.1). Various approaches to statistical sampling have been developed to deal with these issues (e.g. Schulze *et al.*, 1995).

Initially it was thought that a 'black box approach', in which nothing needs to be known about the mechanisms within the box, would be sufficient to model the transfer function that connects input and output fluxes of a system (Jarvis, 1987). However, it has become clear that this is not the case if a predictive capability is required. While it is relatively easy to make predictions about the physico-chemical aspect of fluxes through boundaries, it becomes very difficult to extrapolate across the range of common perturbations, such as drought, if, for example, the process that determines the flux is a hormonal signal from roots to shoots (Schulze, 1994*d*). Thus, the ecological control of fluxes has remained an area of uncertainty because it involves biological regulation, acclimation, adaptation, changing composition of species including replacements and invasion, and interactions between trophic levels.

The most basic method of estimating the magnitude of a flux is to measure the rate of increase of the pool into which it flows, or the rate of decrease in the pool from which it originates. The interpretation of these data is simplified when other fluxes into or out of the pool can be blocked. For example, nitrogen mineralization is generally measured as the rate of increase of the inorganic nitrogen pools, following incubation of a soil sample in the absence of root uptake. This procedure, however, introduces new problems. By sieving soil and extracting roots, the natural soil density and structure are altered, which affects the process to be measured. Thus, it is not generally possible to block a component flux without altering the process under study in other ways.

New approaches to pool and flux observations

In terms of carbon fluxes, Net Primary Productivity (NPP, in most cases defined as seasonal aboveground growth, see also Chapter 7), with its associated flux of nutrients, is the main parameter to be measured in plot studies. Various methods have been developed to assess this quantity, most of them ignoring the important belowground component. The GCTE objective of developing general relations for processes which control fluxes has led to one solution to this problem; based on relations of root distribution with soil depth in different climates (Jackson *et al.*, 1996*b*), predictions of global fine root turnover can be made (Jackson *et al.*, 1997). This is an example of GCTE-initiated research in which global datasets combined with functional relations and consideration of the statistical variation have led to global predictions of parameters which previously were largely unknown.

So far biogeochemical models concentrate on assimilation and respiration of resources in the soil–plant–atmosphere system. They totally neglect the effect and function of herbivory and pests. At this stage there is insufficient detailed understanding of foodweb structures (Pimm, 1993) and their control features for fluxes (Zwölfer & Arnold-Rinehart, 1993) beyond plot scales. This, how-

ever, will be needed for predictions of the effects of global change on pests and herbivores and the consequences for global biogeochemistry (Burdon, 1993; McNaughton, 1993). General overriding relations, as for fine root turnover, are useful also in this case to identify nonlinearities and discontinuities of environmental or system-related parameters on the overall flux. Schulze (1995*a*) has suggested the use of the 'flux control' approach of biochemistry. In this approach ecosystem-level fluxes are related to some key state variables or species diversity, and the slope of the response curve will identify the sensitivity of the process under investigation to that driving variable. Several possible response curves emerge, e.g. in relation to biodiversity, depending on slow- and fast-growing species and the presence or emergence of keystone species effects (Bond, 1993*a*).

In general, the extrapolation of flux measurements of NPP over a season on a single plot to an estimate of the long-term carbon sequestration of the system, although sometimes attempted, is fraught with difficulties. In addition to the complications noted above, there are a number of processes, affecting both ecosystem structure and functioning, operating at larger space and longer time scales which make such extrapolations impossible to do accurately. A recent breakthrough in tackling this problem is the introduction of the concept of Net Biome Productivity (NBP, Schulze & Heimann, 1998), which incorporates flux terms due to disturbances such as harvest by man (e.g. in agriculture and forestry) and fire, and thus provides a biome-level framework for estimating changes in terrestrial pools and fluxes of carbon over long timeframes.

Results and interpretations of measurements of terrestrial carbon pools and fluxes are dealt with in detail in other parts of this book, particularly Chapters 7 and 10 and Sections 12.2.1 and 12.2.2.

2.4.2 Vertical flux measurements

Ecosystem studies require a quantification of the exchange of material across the boundaries, most importantly in a global change context the vertical fluxes between the land surface and the atmosphere. The study of the net exchange of gases between terrestrial ecosystems and the atmosphere has developed rapidly during the past decade, mainly due to the availability of suitable instrumentation (see special issue *Global Change Biology* Vol. 2, No 3, 1996). In this context, ecosystems are often treated as networks of parallel and serial resistors and capacitances (e.g. evaporation from soil surface versus through the plant). There are, however, limitations to the 'big leaf' model of the vegetation canopy (Raupach & Finnigan, 1988) and new models have been developed which avoid this significant simplification (De Pury & Farquhar, 1997). Such models become especially important for the interpretation of mass and isotopic fluxes

between the terrestrial surface and the convective boundary layer (Lloyd *et al.*, 1996).

Despite the emphasis on integrating fluxes over an entire vegetation patch ('footprint' about 1 km^2) by micrometeorological techniques such as eddy covariance, leaf-level parameters such as the response of assimilation to leaf internal CO_2 concentration (Farquhar & Sharkey, 1982) are still required in order to simulate global responses of ecosystems to elevated atmospheric CO_2. The combination of canopy-level gas exchange measurements by eddy covariance and leaf-level measurements has produced response curves of gas exchange and climatic variables which can be used in global models (Leuning *et al.*, 1995). These models can simulate leaf-level responses, such as stomatal closure by air humidity and associated decreases in transpiration, which were not formerly expected to even exist at the canopy level.

Measurements of canopy-level CO_2 exchange give a first estimate of the net balance between assimilatory and respiratory processes of ecosystems. They reveal whether a patch of vegetation is a net carbon source or sink at the time of measurement. Thus, while leaf-level measurements and harvests can quantify NPP, canopy-level eddy covariance measurements quantify Net Ecosystem Exchange (NEE) as a first estimate of Net Ecosystem Productivity (NEP, see also Chapters 7 and 10). Long-term measurements of NEE contain information on climate effects on vertical carbon fluxes, but they do not include major exports of carbon, such as by dissolution in surface or ground water, or by harvest and fire (see Section 2.4.1). Also, by selection of 'uniform stands' (necessary to satisfy the fetch requirements for eddy covariance measurements), canopy-level measurements have not yet dealt with gas-exchange consequences of patchy disturbance, such as tree fall by wind.

A new approach for scaling vertical fluxes from plots to regions has been developed during the past decade, namely the integration of processes at the landscape scale by using daily changes in the Convective Boundary Layer, CBL (Raupach *et al.*, 1992). The CBL can be thought of as a giant cuvette in which gases accumulate over the day, as they cannot escape through the inversion layer at top of the CBL (Lloyd *et al.*, 1996). If the remaining theoretical problems of calculating fluxes from CBL measurements are solved (M.R. Raupach, personal communication), then CBL measurements will allow a quantification of regional fluxes at scales of up to 10 to 50 km^2. Thus, the CBL-derived flux from terrestrial surfaces could include the effects of disturbances and thus would move closer to a measure of NBP (Hollinger *et al.*, 1995; Crawford *et al.*, 1996).

2.4.3 Soil processes and watersheds

Availability of nutrients is a key factor controlling the functioning of terrestrial ecosystems, and is already being strongly affected by global change (see Chapter

1). Although manipulative experiments can help determine the overall system response to changing availability of nutrients (see Sections 2.3.3 and 7.5), observational studies are important in elucidating the mechanisms that affect nutrient uptake and loss by ecosystems.

From earlier studies of the effects of air pollution, soil water solution chemistry emerged as an integrative parameter useful in quantifying nutrient availability. Although relationships determined from solution chemistry appear to hold for most cations, including the toxic effects of aluminium, they do not hold for nitrogen, a key nutrient. Only the nitrate form of nitrogen is mobile in soil water solution. The ammonium form is generally bound to soil or humus particles, and is replaced (mobilized) by a proton in an equilibrium reaction regulated by roots or mycorrhizae. This becomes even more complicated in nitrogen-limited soils, where mycorrhizae and roots of some plant species directly recover organic matter without microbial mineralization (Chapin *et al.*, 1993; Read, 1993). Thus, a low concentration of nitrogen-containing ions in soil solution may express either low availability or high turnover. This creates significant problems for quantifying nitrogen availability in a global change context. For example, through a change in the plant functional type composition of an ecosystem through land-use change, the same soil can be converted from a habitat of high resource availability and turnover (e.g. deciduous forest) into a habitat of low resource availability and turnover (e.g. coniferous forest) (Schulze & Chapin, 1987). Thus, global models that link canopy gas exchange and nitrogen nutrition are based on leaf traits rather than soil characteristics; Lloyd & Farquhar (1996) have made an initial, general attempt to quantify the linkages between soil properties and vegetation growth.

The export of carbon to soils and its immobilization there is a critical component of the terrestrial carbon cycle but remains a big unknown in most flux studies. It is only recently that rooting depth of plants and vegetation types has been reviewed on a global scale (Jackson *et al.*, 1996*a*,*b*; Canadell *et al.*, 1996*a*). Work reported in these reviews shows that roots can be extremely deep (more than 68 m in the Kalahari sands); the carbon turnover rates and contribution to the water and nutrient budgets of the plant by such deep roots remain unknown.

Much of our knowledge about ecosystem-level biogeochemistry comes from watershed studies (e.g. Bormann & Likens, 1979), including their experimental manipulation (e.g. Bayley & Schindler, 1991; Schindler, 1991). The desire to measure all components of the hydrological and biogeochemical balances has led to the use of watersheds as the bounded unit of an ecosystem for such flux studies. The investigation of watersheds may be the only possible way to integrate horizontal biogeochemical fluxes across land surfaces (although one should be aware that any watershed contains a gradient in ecosystems, including

anoxic zones which may buffer and alter the chemistry of the water flow). In the context of global change, the study of watersheds gives insight into element pathways and losses at the plant/soil interface, which is complementary to the vertical flux measurements at the canopy scale (Hornung & Reynolds, 1995).

However, the interpretation of the input/output analysis of watershed biogeochemical studies faces the same problems as the canopy level gas exchange by eddy covariance (Hornung & Reynolds, 1995). The biological regulation inside the 'black box' defined by the catchment influences the output in an unpredictable manner. This is especially true for the nitrogen cycle. For example, the chemical environment in soil macropores differs from the environment in small-diameter soil pores. The watershed chemistry may be driven more by the rapid flow of water through macropores than through fine pores; however, the internal chemistry of the system (e.g. denitrification) may depend strongly on local anoxic zones in soil aggregates (Horn, 1993). In the context of air pollution studies, this made the definition of critical loads difficult where limits were set based on watershed results (Schulze *et al.*, 1989). After sulphur deposition terminated, forests recovered faster than expected. This again illustrates the need to know details of the processes in order to make appropriate predictions, despite the appeal of ecosystem-level flux measurements.

There are other considerations which must be taken into account when using the watershed approach for global change biogeochemical research. Although overall elemental output can be measured accurately, major problems remain with the quantification of inputs because of canopy processes which cannot be detected in the canopy throughfall. Most important is the uptake of N- and S-containing gases through the stomata and their metabolysis in the leaf. It is estimated that the amount of N taken up in the form of NO_x and NH_3 may provide about 25% to 30% of the demand for growth in Central Europe (Schulze, 1994*c*). Furthermore, the heterogeneity of soil often prevents reliable, direct measurements of the ground water flux at a plot scale. As recent techniques have considerably improved the reliability of canopy water flux and tree sapflow measurements, the ground water flux is often estimated by difference from precipitation and canopy water loss (Köstner *et al.*, 1992; Kelliher *et al.*, 1997).

Where an understanding of the impacts of land-use/cover change on horizontal transport processes at larger scales is important, the use of the 'catchment cascades' approach (Fig. 2.4) is a powerful tool for integrating larger regional effects which could either enhance or buffer disturbances. The catchment cascade is based on a gravity-driven chain of mass and energy flows, with the output from one subsystem serving as the input to the next. Measurements of both horizontal and vertical fluxes of important elements are measured at key points along the cascade to identify those points at which elements are lost to the

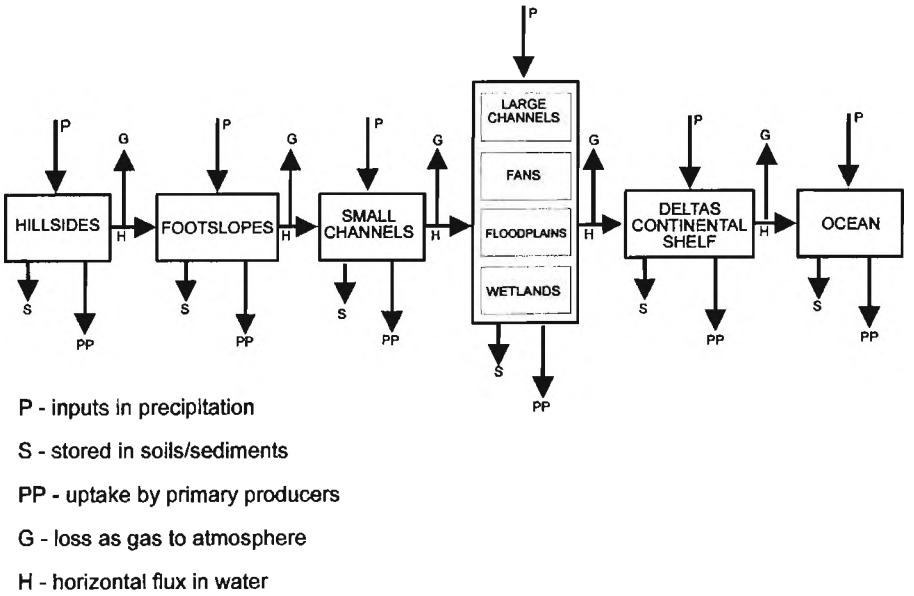

Figure 2.4 A schematic diagram of a 'catchment cascade' approach for estimating both horizontal and vertical fluxes (R.J. Wasson, personal communication).

atmosphere or stored within the catchment. The catchment cascade approach is being used in Southeast Asia to determine the impact of changes in land-use/cover in upland areas on lower lying plains, the coastal zone and the continental shelf area (Lebel Steffen, 1998).

2.4.4 Isotopes

The analysis of naturally occuring stable isotopes (e.g. ^{2}H, ^{13}C, ^{15}N and ^{18}O) is proving increasingly useful in linking ecophysiological processes, ecosystem dynamics and global ecology. The development of this technique has been driven by both the recognition that the isotopic composition of a material contains information about its source as well as the ecological processes that lead to its formation, and by technological improvements that allow samples to be measured more rapidly and accurately than before (Rundel *et al.*, 1989; Ehleringer *et al.*, 1993). Stable isotope analysis constrains uncertainties about source/sink relationships within ecosystems; it allows a separation of net mass flow and one-way fluxes (Lloyd *et al.*, 1996); and stable isotopes can be applied as tracers (e.g. Buchmann *et al.*, 1995).

Fractionation events associated with ecophysiological processes often scale to, and have significant impacts on, processes at the ecosystem level. Carbon isotope ratios and the resulting differences in discrimination by C3 and C4

canopies permit scaling of ecophysiological processes to the canopy level, which allowed Lloyd & Farquhar (1994) to estimate the global impacts of C3 and C4 ecosystems on biosphere–atmosphere gas exchange. At the ecophysiological level the carbon isotope ratio of C3 plants varies with the ratio of assimilation to leaf conductance (Farquhar *et al.*, 1989), which leads to differences in the ratio of CO_2 to water vapour exchange at the ecosystem level (Farquhar & Lloyd, 1993). More recently, CO_2 ratios have been used to separate respiration from the one-way fluxes of CO_2 into and out of canopies (Lloyd *et al.*, 1996).

The abundance of carbon and oxygen isotopes in CO_2 varies significantly on both a seasonal and a latitudinal basis, reflecting the annual dynamics of productivity and decomposition in different ecosystems (Francey & Tans, 1987). Farquhar *et al.* (1993) developed a mechanistic model showing that the biosphere should have a substantial impact in modifying the ^{18}O content of atmospheric CO_2 and that terrestrial systems differed from marine systems in their impact on the ^{18}O signal (Keeling *et al.*, 1996*b*). Ciais *et al.* (1997) combined this model with global inverse modelling approaches to confirm that terrestrial ecosystems (and not marine ecosystems) are responsible for the missing carbon sink (see Chapters 10 and 12). This has been quantified by Keeling *et al.* (1996*b*) based on isotope measurements in conjunction with long-term measurements of O_2/N_2 ratios.

Deuterium and oxygen isotopes in water are proving to be equally useful tools for understanding the water-related dynamics of ecosystems (Dawson & Ehleringer, 1991; Ehleringer & Dawson, 1992). Plants do not discriminate during water uptake, allowing patterns of water uptake and loss by different ecosystem components (Schulze *et al.*, 1996*a*) to be quantified. These approaches show that not all ecosystem components respond equally to variations in winter–summer precipitation events (Ehleringer *et al.*, 1991) and, in some cases, long-held views on the sources of water for trees have had to be modified (Dawson & Ehleringer, 1991; Thornburn *et al.*, 1992). At the ecosystem level, isotopic analysis of atmospheric water vapour allows the impact of transpirational fluxes on regional processes to be constrained (Brunnel *et al.*, 1991).

Over the past several years there has been substantial progress in isotope ratio mass spectrometer instrumentation. The capacity to analyse isotopic composition rapidly and the capability to measure small samples have reshaped sampling approaches. Carbon and nitrogen isotope ratios of organic materials are now routinely analysed by linking elemental analysers to mass spectrometers (Gebauer & Schulze, 1991). An automated approach allows 75 or more samples per day to be routinely measured. Linking continuous-flow mass spectrometry and gas chromatography permits even faster analysis of organic material and allows for high-precision analysis of smaller samples, including atmospheric and trace gases. Equally important has been the use of tracers, especially ^{15}N, to

quantify ecosystem internal fluxes and identify mechanisms of turnover (Davidson *et al.*, 1992).

2.4.5 Remote sensing and geographical information systems

Remotely sensed data, particularly when manipulated in Geographic Information Systems (GIS), have given ecologists a tool to undertake spatially integrated analysis at regional scale. The regular and frequent overpass of sensors carried on satellite platforms allows consistent repeated observations of a large number of locations. At the most basic level, it has allowed the cost-effective mapping of spatial patterns over large areas. The comparison of maps for periods a decade apart provided an accurate way to determine land-cover changes (e.g. Skole & Tucker, 1993). The characteristics of the sensors has allowed them to be calibrated to important ecosystem variables, such as green leaf cover (Field *et al.*, 1993) or the detection of fires (Scholes *et al.*, 1996*b*). Chapter 5 discusses global databases in more detail.

2.5 Conclusions

Technological advances, especially in the last three decades, have provided powerful new tools for ecosystem analysis. These include:

- micrometeorological and isotopic techniques for estimating the flux of matter and energy within ecosystems, and between ecosystems and the atmosphere, geosphere and biosphere;
- techniques for altering selected environmental factors such as CO_2 and temperature at the plot scale;
- remote sensing to provide detailed spatial and temporal data over large areas;
- statistical and numerical modelling techniques capable of analysing multivariate, nonlinear problems;

The value of the information which can be obtained from studies on small plots can be greatly enhanced by:

- subjecting the whole plot to manipulations, such as elevated CO_2, fertilizer, irrigation or warming, especially if response surfaces and not only factorial combinations are obtained;
- consistent observation over long periods of time;
- comparison of similar experimental treatments in different parts of the world (transects, see Chapter 4);
- using 'natural experiments' provided by gradients of environmental drivers.

A number of parameters and processes remain uncertain and will need further investigations for their quantification. The most important of these include:

- fluxes in the soil;
- a quantification of resource availability for the vegetation cover.