A search for predictive understanding of plant responses to elevated \[\text{CO}_2\]

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Abstract

This paper reviews two decades of effort by the scientific community in a search for predictive understanding of plant responses to elevated \[\text{CO}_2\]. To evaluate the progress of research in leaf photosynthesis, plant respiration, root nutrient uptake, and carbon partitioning, we divided scientific activities into four phases: (I) initial assessments derived from our existing knowledge base to provide frameworks for experimental studies; (II) experimental tests of the initial assessments; (III) in cases where assessments were invalidated, synthesis of experimental results to stimulate alternative hypotheses and further experimentation; and (IV) formation of new knowledge. This paper suggests that photosynthetic research may have gone through all four phases, considering that (a) variable responses of photosynthesis to \[\text{CO}_2\] are generally explainable, (b) extrapolation of leaf-level studies to the global scale has been examined, and (c) molecular studies are under way. Investigation of plant respiratory responses to \[\text{CO}_2\] has reached the third phase: experimental results have been accumulated, and mechanistic approaches are being developed to examine alternative hypotheses in search for new concepts and/or new quantitative frameworks to understand respiratory responses to elevated \[\text{CO}_2\]. The study of nutrient uptake kinetics is still in the second phase: experimental evidence has contradicted some of the initial assessments, and more experimental studies need to be designed before generalizations can be made. It is quite unfortunate that we have not made much progress in understanding mechanisms of carbon partitioning during the past two decades. This is due in part to the fact that some of the holistic theories, such as functional balance and optimality, have not evolved into testable hypotheses to guide experimental studies. This paper urges modelers to play an increasing role in plant–\[\text{CO}_2\] research by disassembling these existing theories into hypotheses and urges experimentalists to design experiments to examine these holistic concepts.

Keywords: acclimation, carbon dioxide, carbon partitioning, global change, nutrient uptake, photosynthesis, respiration, science philosophy.

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Introduction

It has been more than 20 years since the US Department of Energy (DOE) sponsored a workshop in Miami, Florida in 1977 which officially incorporated plants and terrestrial ecosystems into its Carbon Dioxide Research Program (Elliot & Machta 1979). Since the 1977 DOE workshop, predictive understanding of plant responses to rising atmospheric \[\text{CO}_2\] has been sought in order to help explain the past and to forecast future changes in global carbon cycling. The need for plant and ecosystem studies was stimulated largely by modelling results that had long suggested that the global carbon budget could not be balanced without storage of carbon in terrestrial ecosystems (Bacastow & Keeling 1973). Recently, more evidence has been presented to support the idea that terrestrial ecosystems play a critical role in modulating carbon balance in the earth system (Tans et al. 1990, 1993; Lloyd & Farquhar 1994; Ciais et al. 1995). To provide explanatory and predictive understanding of rising atmospheric \[\text{CO}_2\], the study of plant and ecosystem responses to elevated \[\text{CO}_2\] is necessary.

Stimulated by the global carbon cycling issue, signific-
plant research has been carried out by the international scientific community on many aspects of plant biology, including photosynthesis, respiration, nutrient uptake, and carbon partitioning. By 1993, ≈ 1500 papers had been published on plant responses to elevated \( [CO_2] \) (Körner 1993). In 1996 alone, nearly three hundred research papers were published in this area. Plant–CO2 research is booming because atmospheric \( [CO_2] \) is increasing at an unprecedented rate and also because an increase in \( [CO_2] \) can stimulate horticultural and agronomic yields (Acock & Allen 1985) and alter plant and ecosystem function and structure in the earth system (Mooney et al. 1991). In addition, plant–CO2 research in the past two decades has provided great opportunities to advance our understanding of many basic biological processes (Wardlaw 1990). As a result, many of the existing paradigms in plant biology have been re-examined and expanded in the context of global environmental change (Bazzaz 1990; Field et al. 1992).

These intensive studies of plant responses to elevated \( [CO_2] \) have rapidly broadened the scope of plant biology. The research scope presently ranges in scale from molecular response to \( [CO_2] \) (Griffin & Seemann 1996) to \( CO_2 \) impact on species diversity in ecosystem communities (Bazzaz 1990) and global biosphere fluxes (Melillo et al. 1993). Growth of plants in elevated \( [CO_2] \) usually results in accumulation of leaf starch and soluble sugars, and photosynthetic acclimation. Molecular studies have examined sugar-regulated gene expression as well as functions of encoded proteins and associated metabolic fluxes (Stitt 1991; Van Oosten et al. 1994). At the global scale, experimental results have been integrated into global biosphere models to predict potential changes in terrestrial ecosystems in the next century when atmospheric \( [CO_2] \) gradually increases to the elevated level (e.g. Polglase & Wang 1992).

While many aspects of \( CO_2 \) research have developed quickly, some fundamental plant processes are extremely variable in response to elevated \( [CO_2] \). For example, the apparent respiration rate increases for some species and decreases for others during either short-term exposure to or long-term growth in elevated \( [CO_2] \) (Amthor 1997). Similarly, diverse responses of nitrogen uptake, stomatal conductance, carbon allocation (e.g. root/shoot ratio), and photosynthetic capacity to \( [CO_2] \) have been observed. Many of these diverse responses are presently not explicable, hindering utilization of experimental results and impeding integration of plant biology into global studies.

This paper will review the process of scientific investigation in plant–CO2 research, focusing on activities playing critical roles in advancing our predictive understanding of plant responses to elevated \( [CO_2] \). Toward that end, we distinguish four phases of scientific activities in knowledge advancement: (I) initial assessments derived from our existing knowledge base to provide frameworks for experimental studies; (II) experimentation to accept or reject the initial assessments; (III) in cases where assessments proved to be inaccurate, synthesis of experimental data to stimulate alternative hypotheses and further experimental studies; and (IV) formation of new knowledge. We will first provide an overview of the four phases and then discuss each phase in relation to studies of photosynthesis, respiration, nutrient uptake, and carbon partitioning in response to elevated \( [CO_2] \).

While this paper is primarily organized through the four phases, the evolution of research is described in three tables on individual processes of photosynthesis, respiration, and carbon partitioning. Research activities on other subjects (e.g. stomatal conductance and plant competition) can be similarly evaluated by the four-phase approach. In addition, this paper revolves around experimental studies leading to predictive understanding. Other aspects of \( CO_2 \) research have been extensively reviewed in other papers (e.g. Bazzaz 1990; Allen et al. 1992; Field et al. 1992; Körner 1995; Reynolds et al. 1996; Amthor 1997; Drake et al. 1997).

Overview of four phases of plant–CO2 research

As in most scientific disciplines, research on plant responses to elevated \( [CO_2] \) has undergone four distinguishable phases of progress (Fig. 1). The core knowledge base from studies of plant responses to soil fertility (e.g. nitrogen and phosphorus) and light availability provides a starting point in the first phase. Initial assessments of possible responses of plants to elevated \( [CO_2] \) are imposed in order to frame experimental studies (Field et al. 1992). For example, a biochemically based model developed by Farquhar et al. (1980) that describes photosynthetic sensitivity to \( [CO_2] \) has been used to assess possible changes in photosynthetic capacity in response to elevated \( [CO_2] \) (Pearcy & Bjorkman 1983). Based on the colimitation concept in the model, it was inferred that plant growth in elevated \( [CO_2] \) should result in relative reduction of rubisco (ribulose-1,5-bisphosphate carboxylase-oxygenase) capacity and enhancement of RuBP (ribulose bisphosphate) regeneration (Sage 1990). To assess possible changes in carbon partitioning, a functional balance concept developed by Brouwer (1962) was used. Since nitrogen fertilization enhances root activities, leading to reduced root/shoot ratio (Wilson 1988), it seemed logical to expect that the root/shoot ratio would be increased in elevated \( [CO_2] \) because \( CO_2 \) enhances the shoot activities.

In Phase II of plant–CO2 research, experiments are designed to test the hypotheses derived from the initial assessments. Based on experimental results, some of these assessments are accepted and some are rejected. For
PLANT RESPONSES TO [CO₂] vary widely, a search for predictive understanding of the variable responses has been undertaken through data synthesis. Many synthetic papers have been written on plant responses to elevated [CO₂] (e.g. Kimball 1983; Poorter 1993; Gunderson & Wullschleger 1994; Rogers et al. 1994; Sage 1994; Curtis 1996). Curtis (1996) recently introduced a meta-analysis method to provide statistically unbiased estimates of mean responses of experimental data from multiple projects. Synthesis, direct experimental evidence, or both may lead to alternative hypotheses. On photosynthetic acclimation to elevated [CO₂], for instance, at least several hypotheses have been proposed (e.g. disruption of chloroplast, end-product inhibition through sugar accumulation, and increased mesophyll growth). These hypotheses stimulate more experimental studies.

The hypothesis-experiment cycle may be iterated several times. Eventually in Phase IV, this scientific process leads to development of new knowledge regarding plant responses to elevated [CO₂]. For example, the canopy productivity index (i.e. the annual production of wood per unit of leaf area) has been found to be similar across several broadleaf tree species despite the fact that CO₂ stimulation of tree biomass growth is extremely variable (Norby 1996). Molecular studies have recently led to the conclusion that the accumulation of leaf hexoses initiates a chain of molecular and biochemical responses that may be directly related to acclimative changes in rubisco activities and mRNA transcripts when plants are exposed to elevated [CO₂] (Sheen 1994; Van Oosten & Besford 1994).

Initial assessments
As discussed, initial assessments are made from the existing knowledge base. The core knowledge base is generally organized by various frameworks which influence our thinking (i.e. schools of thought). The knowledge base of plant biology is fairly rich and consists of many schools of thought including: (i) plant functional balance (Brouwer 1962; Reynolds & Thornley 1982; Wilson 1988); (ii) resource balance (Field et al. 1992); (iii) plant functional types (Chapin 1991); (iv) ontogenetic drift (Coleman et al. 1993); (v) growth analysis (Bazzaz 1993); (vi) nitrogen productivity (Ågren 1985; Ågren & Ingstad 1987); (vii) source-sink regulation (Stitt 1991); and (viii) optimality (Bloom et al. 1985; Hilbert 1990). For example, the growth analysis approach separates plant growth rate into several components, including net assimilation rate per unit of leaf area, leaf area ratio, and specific leaf area. These components have been used to assess the impact of global change on plant growth and allocation (Bazzaz 1993). These schools of thought identified above are not mutually exclusive. Each has its own features and differentially

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influences the thinking of various research groups, though.

These schools of thought have not equally influenced research into plant responses to elevated [CO₂]. In the history of CO₂ research, the knowledge base related to growth analysis has been one of the most influential in directing experimental studies. Based on the fact that rising atmospheric [CO₂] will directly increase photosynthetic carbon fixation and reduce water loss through transpiration, Strain & Cure (1985) proposed three-level (primary, secondary, and tertiary) effects of elevated [CO₂] on plants. The primary effects are those of [CO₂] on photosynthesis, transpiration, and stomatal conductance. These physiological processes are directly responsive to changes in ambient [CO₂]. The secondary effects are on primary productivity, growth, and carbon partitioning which are influenced by [CO₂] through altered carbohydrate availability and plant water status. The tertiary effects are on secondary chemical compounds that regulate herbivory and then community dynamics. Influenced by this framework, a considerable fraction of the plant-CO₂ research has focused on biomass and gas exchange, providing a rich array of data for developing plant growth models (Reynolds et al. 1996). Other influential frameworks include those derived from the concept of source–sink regulation by Stitt (1991) and Farrar & Williams (1991) as well as the resource-based approach and plant functional types by Mooney et al. (1991). The source–sink framework is particularly useful in guiding molecular and biochemical studies of plant responses to [CO₂]. The resource-based approach provides a research framework in relation to an array of ecosystem types ordinated by two axes of drought stress and nutrient availability. This framework is presently influencing ecosystem-scale studies using Free-Air CO₂ Enrichment (FACE) facilities and open-top chambers.

Initial assessments become useful in guiding experimental studies only if testable hypotheses can be derived from the core knowledge. For example, the paper by Stitt (1991) disassembled the source–sink concept into a variety of testable hypotheses. The paper by Mooney et al. (1991) placed the resource balance and functional type concepts in a context of different ecosystem types. In contrast, other concepts such as optimality, nitrogen productivity, and functional balance have not become major frameworks in influencing experimental studies. These concepts have been integrated into modelling studies (Reynolds & Thornley 1982; Ågren & Ingestad 1987; Hilbert & Reynolds 1991) and have proved useful in interpreting whole-plant responses to environmental factors (Bloom et al. 1985). These holistic approaches have, unfortunately, not been well disseminated as testable hypotheses that experimentalists can easily test. It is the future responsibility of the CO₂ research community to explore how to design experiments to measure, for example, marginal cost vs. marginal benefit associated with environmental changes.

**Experimental tests of initial assessments**

Many of the initial assessments of physiological processes in response to elevated [CO₂] have not been confirmed by experimental evidence. For example, the speculation in the early 1980s that photosynthetic capacity should decrease in elevated [CO₂] (Pearcy & Bjorkman 1983) was gradually challenged by experimental data (Table 1). The photosynthetic capacity of plants grown in elevated [CO₂] has been experimentally shown to increase, decrease, or not change depending on species and growth environments (Campbell et al. 1988; Sage et al. 1989). For example, a comparative study with five species conducted by Sage et al. (1989) demonstrated that growth in elevated [CO₂] led to either an increase or decrease in maximal values as well as initial slopes of A/Ci (assimilation/intercellular [CO₂]) responses.

The experimental evidence has also challenged some of the existing assumptions on respiratory responses to elevated [CO₂]. According to the respiratory control theory, the rate of respiration is primarily regulated by consumption of respiratory products (e.g. ATP, NAD(P)H) and carbon skeleton intermediates) (Palmer 1984; Ap Rees & Williams 1990; Amthor 1991). Since the consumption of respiratory products is associated primarily with growth and maintenance processes, CO₂-enhanced plant growth should require more respiratory products, leading to a high respiration rate. However, experimental results suggest that apparent respiration rates can either decrease (Amthor et al. 1992; Bunce 1990; Thomas & Griffin 1993) or increase (Ryle et al. 1992a,b; Ziska & Bunce 1993) during short-term CO₂ increases. Long-term growth of plants in elevated [CO₂] also leads to variable responses to respiration (Poorter et al. 1992; Thomas & Griffin 1993; Wullschleger et al. 1994; Amthor 1997). The variable long-term responses of respiration may still be consistent with the control theory because of the interactive effects of increased growth and decreased tissue nitrogen concentration on respiration (Amthor 1997). However, the mechanisms of short-term effects of [CO₂] on respiration are unclear and may be related to inhibition of cytochrome oxidase activity (González-Meler et al. 1996) (Table 2).

Many of the existing notions on nutrient uptake cannot be supported by experimental results to date, either. Plant nutrient acquisition is expected to be enhanced when nutrient demand associated with growth is accelerated in elevated [CO₂]. Measured phosphorus uptake, however, did not show an increase in either the uptake rate or root fraction (Bassirirad et al. 1996a). Similar physiological rates of ammonium uptake were unchanged.
Table 1 Evolution of the research on photosynthetic responses to elevated [CO2] characterized by four phases: Phase I is the initial assessment of photosynthetic responses to elevated [CO2]; Phase II is experimental tests of the initial assessments; Phase III is generation of alternative hypotheses; and Phase IV is the formation of new knowledge. (See text for more description of the phases.)

<table>
<thead>
<tr>
<th>Phase</th>
<th>Description</th>
<th>References</th>
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| I     | 1. Photosynthetic rate will be increased.  
2. Photosynthetic capacity will be decreased (i.e. downregulation). | Pearcy & Bjorkman (1983) |
| II    | Experimental data confirmed the increase in photosynthetic rate but rejected the speculation that photosynthetic capacity is always decreased. | Sage et al. (1989) |
| III   | Hypotheses to explain responses of photosynthetic capacity to [CO2] include:  
1. Chloroplast breakdown;  
2. N redistribution;  
3. Source-sink regulation;  
4. End-product inhibition;  
5. Starch accumulation;  
6. Morphological growth;  
7. Compensatory changes in N dilution and mesophyll growth. | DeLucia et al. (1985)  
Sage (1994)  
Stitt (1991)  
Long (1991)  
Long & Drake (1992)  
Vu et al. (1989)  
Luo et al. (1994) |
| IV    | 1. The balance between biochemical downregulation and morphological upregulation explains both up- and down-regulation of photosynthetic capacity.  
2. The accumulation of leaf hexoses initiates a signal-response mechanism that represses transcription of many photosynthetic genes.  
3. Photosynthetic sensitivity is scaleable to the globe but the acclimative changes are not. | Luo et al. (1994)  
Jang & Sheen (1994)  
Luo et al. (1996) |

Table 2. Evolution of the research on respiratory responses to elevated [CO2] characterized by four phases. (See Table 1 and text for description of the four phases.)

<table>
<thead>
<tr>
<th>Phase</th>
<th>Description</th>
<th>References</th>
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| I     | 1. Whole-plant growth and maintenance respiration will increase because of more growth and large plants.  
2. Growth cost and specific maintenance respiration rate may decrease if tissue composition changes.  
3. No prediction prior to experiments was made on short-term CO2 effect. | Amthor (1991) |
| II    | 1. Experimental data generally confirmed that whole-plant growth and maintenance respiration increases and growth cost and specific maintenance respiration rates decrease.  
González-Meler et al. (1996)  
Amthor et al. (1992) |
| III   | Hypotheses to explain the short-term CO2 effect on respiration include:  
1. Measurement error with leaking chambers;  
2. Inhibition of enzymatic activities;  
3. Dark CO2 fixation;  
González-Meler et al. (1996)  
Amthor (1995)  
Palet et al. (1992) |
| IV    | 1. The long-term CO2 effects on whole-plant growth and maintenance respiration, growth cost, and specific maintenance respiration are conceptually explicable and yet to be evaluated quantitatively.  
2. Inhibition of cytochrome c oxidase activity may be partly responsible for the short-term CO2 effects. | Amthor (1997)  
González-Meler et al. (1996)  
Azcón-Bieto et al. (1994) |

with elevated [CO2] and rates of nitrate uptake decreased in a study of six species in California grasslands (Jackson & Reynolds 1996). Opposite to that finding are the nutrient uptake rates by field-grown loblolly pine saplings: [CO2] enrichment enhanced the root uptake capacity for nitrate but not for ammonium (Bassirirad et al. 1996b). Yet the additional nutrient demand can be balanced by increasing root uptake areas (Jackson & Reynolds 1996) and/or reduced tissue nutrient concentration in elevated [CO2].

Rigorous experimental tests of existing concepts on carbon partitioning have rarely been conducted (but see Chu et al. 1992) although many experiments observed simple indices such as root/shoot ratio and leaf area ratio (Rogers et al. 1994) (Table 3). These simple indices do not...
Table 3 Evolution of the research on carbon partitioning responses to elevated [CO₂] characterized by four phases. (See Table 1 and text for description of the four phases.)

<table>
<thead>
<tr>
<th>Phase</th>
<th>Description</th>
<th>References</th>
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<tbody>
<tr>
<td>I</td>
<td>Root/shoot ratio is speculated to increase because of enhancement in shoot activity</td>
<td>Wilson (1988)</td>
</tr>
<tr>
<td>II</td>
<td>Experimental data indicate root/shoot ratio increases for some species and decreases for others, rejecting the initial hypothesis.</td>
<td>Rogers et al. (1994)</td>
</tr>
<tr>
<td>III</td>
<td>Hypotheses that possibly explain the diverse carbon partitioning include:</td>
<td>Hilbert (1990)</td>
</tr>
<tr>
<td></td>
<td>1. Optimality;</td>
<td>Luo et al. (1994)</td>
</tr>
<tr>
<td></td>
<td>2. Growth/photosynthesis balance;</td>
<td>Coleman et al. (1993)</td>
</tr>
<tr>
<td></td>
<td>3. Ontogenetic drift;</td>
<td>Reynolds &amp; Thornley (1982)</td>
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<tr>
<td></td>
<td>4. Functional balance;</td>
<td>Reynolds &amp; Chen (1996)</td>
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<tr>
<td></td>
<td>5. Coordination;</td>
<td>Stitt (1991)</td>
</tr>
<tr>
<td></td>
<td>Variation in root/shoot ratio may be explicable by increased nitrogen productivity and reduced carbon use efficiency for carbon fixation and nutrient uptake. Critical datasets are required to test the above hypotheses.</td>
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reveal much about the mechanisms of carbon partitioning. To test the functional balance concept, for example, specific shoot and root activities (i.e. mass-based, whole-shoot photosynthetic rate and whole-root nutrient or water uptake rates) and shoot and root biomass should be simultaneously measured (Wilson 1988). To the best of our knowledge, no single experiment has been undertaken to measure these components in order to test the functional balance concept in elevated [CO₂]. The root/shoot ratio alone cannot validate or falsify the concept because the balance between shoot with root functioning also depends on specific root and shoot activities. These activities have been shown to vary with elevated [CO₂]. In addition, a modelling study by Luo et al. (1994) has demonstrated that two partitioning schemes (nitrogen productivity and functional balance) lead to contradictory predictions on the effects of nitrogen on the root/shoot ratio. A model formulation based on nitrogen productivity suggests root/shoot ratio should increase in elevated [CO₂] with decreased plant nitrogen concentration. The opposite is suggested by the functional balance. It remains a challenge for experimentalists to design tests of these concepts.

Synthesis and alternative hypothesis

A great amount of experimental data have been accumulated since 1977. Because experimental results have proven to be highly variable and often species- and condition-specific, synthesizing experimental results across species and growth environments has become a very effective approach to generalizing plant responses to elevated [CO₂]. The synthetic papers can be grouped into two types: (i) single variable and (ii) multiple variables with a central theme. For example, papers by Kimball (1983), Poorter (1993), Gunderson & Wullschlegel (1994), and Amthor (1997) synthesized published data on crop yield, biomass growth, photosynthesis, and respiration in response to elevated [CO₂], respectively. One of the primary goals of these synthetic papers has been to provide mean responses and variability, usually associated with different types of plants (e.g. C3 vs. C4 plants). The synthetic papers dealing with multiple variables generally attempt to explain observed phenomena by integrating several underlying processes having variable responses (e.g. Long 1991; McMurtrie & Wang 1993; Luo et al. 1994; Poorter et al. 1997). For example, Long (1991) synthesized the responses of starch content, soluble sugars, transpiration, and rubisco content to elevated [CO₂] and incorporated them into a model to predict photosynthetic responses to elevated [CO₂]. Poorter et al. (1997) integrated the leaf chemical composition of 27 species in an attempt to understand CO₂ effects on tissue construction costs and growth respiration. In order to explain nonlinear photosynthetic responses to [CO₂], Luo et al. (1998) have examined relationships among [CO₂], photosynthesis, nitrogen, and specific leaf area by synthesizing data from several CO₂ projects.

Experimental results and/or their synthesis often lead to alternative hypotheses and further experimentation. In searching for mechanisms to explain the diverse photosynthetic responses to long-term growth in elevated [CO₂], for example, several hypotheses have been proposed (Table 1). Among them are (i) source-sink limitation (Farrar & Williams 1991; Stitt 1991), (ii) chloroplast breakdown by oversize starch grains (Delucia et al. 1987), (iii) starch accumulation inhibition (Long & Drake 1991), (iv) phosphorus limitation (Sage 1990, 1994; Harley et al. 1992), (v) pot-size effect (Arp 1991; Thomas & Straeher 1991), (vi) morphological changes, (Vu et al. 1989; Si...
et al. 1998), and (vii) balance between biochemical downregulation and morphological upregulation (Luo et al. 1994, 1998). Chloroplast breakdown has not been observed in many subsequent experiments. An increase in leaf soluble carbohydrate has been consistently observed in many plants grown at elevated [CO₂] (Long & Drake 1992), but the increase is not always well correlated with changes in photosynthetic capacity (Bunce et al. 1995; Jacob et al. 1995). End-product inhibition, phosphorus limitation, and source-sink regulation have been modelled (Hilbert et al. 1991; Long 1991; Harley et al. 1992) and found to partly explain the downregulation but not the upregulation. Luo et al. (1994) evaluated various hypotheses in a quantitative framework and concluded that a balance between biochemical downregulation and morphological upregulation can account for most of the observed variability in photosynthetic acclimation to elevated [CO₂]. The biochemical downregulation encompasses various processes including CO₂-induced inorganic phosphorus limitation, end-product inhibition through nonstructural carbohydrate (e.g. sugar), depressed gene expression, and reduced rubisco amount and/or activity. These biochemical processes generally result in lower photosynthetic rate and capacity (i.e. downregulation). The morphological upregulation is CO₂-stimulated mesophyll tissue growth, possibly including expanded cell volumes and cell layers in a leaf. The balance hypothesis was tested by manipulative experiments in the field (Jackson et al. 1995) and in a controlled environment (D.A. Sims and Y. Luo, unpubl. data).

Respiration in response to both short-term exposure or long-term growth in elevated [CO₂] has also been extensively studied (Burce 1990; Poorter et al. 1992; Wullschleger et al. 1994; Amthor 1995). The review by Poorter et al. (1992), for example, found that apparent leaf respiration rates increased by an average of 16% on a leaf area basis but decreased by an average of 14% on a leaf mass basis for plants grown in elevated [CO₂]. Alternative hypotheses on the variable, long-term effects of [CO₂] on respiration include reduction in mitochondrial enzyme activities and concentration (Azcón-Bieto et al. 1994), regulation of respiration by tissue carbohydrate concentration (Farrar & Williams 1991), changes associated with increased photosynthesis and growth (Amthor 1997). While those hypotheses seem adequate to explain and predict respiratory responses to long-term exposure to elevated [CO₂], a quantitative evaluation against concomitant measurements of growth, tissue chemical composition, and respiration is urgently needed to provide insights into respiratory physiology.

A short-term increase in [CO₂] from ≈ 350 to 700 ppm (in the dark) has been found to reduce leaf respiration by 10–30% across 37 species (Amthor 1997). Several hypotheses have been proposed to identify the possible mechanisms causing this short-term response (Table 2). González-Meler et al. (1996) suggests that inhibition of cytochrome c oxidase and succinate dehydrogenase may be a mechanism of the short-term depression. Other possible mechanisms causing the short-term reduction in respiration include dark [CO₂] fixation by several nonphotosynthetic carboxylases (Amthor 1995), instrumental inaccuracy (Amthor 1997), and substrate stimulation of alternative pathway (Palet et al. 1992). Tests of these hypotheses are under way and will duly advance our understanding of the basic mechanisms of respiratory response to elevated [CO₂].

Only a few studies have been reported on the kinetics of root nutrient uptake with respect to elevated [CO₂] (Bassirirad et al. 1996a,b; Jackson & Reynolds 1996). The general trend of nutrient uptake in response to elevated [CO₂] will not be clear until more experiments are completed involving a variety of species. Although many data are available on plant growth components, it is not easy to assemble the data sets necessary for testing hypotheses on carbon partitioning, largely because the data were not collected under guidance of synthetic frameworks. Nonetheless, the extremely large database accumulated in the past 20 years on many aspects of plant responses to elevated [CO₂] may provide an opportunity for further synthetic studies and modelling integration. Modelling studies, in combination with data synthesis and hypothesis testing, may become a very fruitful approach to improved understanding of the CO₂ effects.

New knowledge

New knowledge can be evaluated using several criteria including (i) phenomenological consistency of plant responses, (ii) explanatory power of observed phenomena, (iii) mechanistic understanding of plant processes, and (iv) scalability of our knowledge across spatial and temporal scales. One notable consistency is that the majority of plant processes in response to elevated [CO₂] are extremely variable, increasing for some species grown in certain environments but decreasing for other species grown in different environments. Consistent responses to elevated [CO₂] include increased photosynthetic rate (Gunderson & Wullschleger 1994; Curtis 1996), increased nonstructural carbohydrate concentration (Long 1991), increased nitrogen and water-use efficiency (Field et al. 1995; Drake et al. 1997), and decreased leaf and plant nutrient concentration (Luo et al. 1994). While there are some exceptions, it appears to be the pattern that respiration rates increase for crop species and decrease for perennial native species grown in high [CO₂] (González-Meler et al. 1996).
The explanatory power of observed phenomena lies in providing causal relationships between the phenomenon in question and underlying processes. Variable responses of photosynthetic capacity to elevated \([\text{CO}_2]\) can be explained largely by a balance of biochemical down-regulation and morphological up-regulation (Luo et al. 1994; 1998) (Fig. 2). When growth at elevated \([\text{CO}_2]\) leads to morphological up-regulation that is larger than decreases in the biochemical capacity of photosynthesis, photosynthesis is up-regulated. Otherwise, photosynthesis is down-regulated (Table 1). Variation of the root/shoot ratio in response to elevated \([\text{CO}_2]\) may be explained by increased nitrogen productivity and reduced carbon use efficiency for carbon fixation and nutrient uptake (Luo et al. 1994). Variable patterns in tissue construction cost and growth respiration are predictable from \(\text{CO}_2\)-induced variation in tissue chemistry. A 42% increase in nonstructural carbohydrate and 9% decrease in protein account for most of the 3% decrease in construction cost and 11% decrease in growth respiration (Poorter et al. 1997).

Mechanistic studies have been conducted on photosynthetic responses to \([\text{CO}_2]\) at molecular and biochemical levels. Increased carbohydrate production at elevated \([\text{CO}_2]\) may exceed the demand from sinks (Stitt 1989) leading to depression of gene expression (Nie et al. 1995a,b) and reduction in mRNA contents (Krapp et al. 1993; Jang & Sheen 1994; Van Oosten & Besford 1994; Van Oosten et al. 1994) and photosynthetic enzymes (Besford et al. 1990; Krapp et al. 1991; Socias et al. 1995). The recent molecular studies have suggested that upregulated gene expression is responsible for change photosynthetic proteins during acclimation to elevated \([\text{CO}_2]\). In particular, Jang & Sheen (1994) have proposed that the accumulation of leaf hexoses can initiate signal-response mechanism that involves cytoplasmic hexokinase and ultimately represses transcription many photosynthetic genes, including rubisco (Table 1). Continued advancement in molecular studies will help reveal the fundamental mechanisms of photosynthetic acclimation to elevated \([\text{CO}_2]\).

To date, many results of plant-level studies have been integrated into regional and global models. For example, the photosynthetic model by Farquhar et al. (1980) for the growth \(\beta\) factor (Wullschleger et al. 1995; Amthor...
has argued for a functional convergence hypothesis to search for scaleable physiological parameters. Field (1991) has suggested that light-use efficiency (defined as the ratio of dry matter production to the integrated energy absorbed) is a reasonable predictor of net primary productivity at large scales in combination with remote sensing data. In addition, Polglase & Wang (1992) and Luo & Mooney (1996) have recently examined leaf photosynthetic sensitivity to an increase in [CO$_2$] (normalized photosynthetic response to a small increase in [CO$_2$]) and found that it is independent of interspecific variation and growth environments for C3 plants and only varies with measurement temperature. This independence has the potential to simplify global extrapolation of leaf-level studies to provide a baseline prediction of a marginal increment in global carbon influx stimulated by a marginal increase in atmospheric [CO$_2$] (Luo et al. 1996). However, the actual increment in global carbon influx as stimulated by rising atmospheric [CO$_2$] has to account for aclimative changes in leaf, plant, canopy scales, temporal shifting of growing seasons, and biome movement (Luo & Mooney 1995).

Concluding remarks

Current understanding of plant responses to [CO$_2$]

Extensive research on photosynthetic acclimation to [CO$_2$] has been conducted. Although variable photosynthetic responses to [CO$_2$] are generally explainable by interactive changes in leaf biochemistry and morphology, predicting acclimation of different species in natural ecosystems is still challenging. Extrapolation of leaf-level studies to larger scales depends on improved understanding of canopy development and community dynamics in elevated [CO$_2$]. We are hopeful that molecular studies may improve our fundamental knowledge of photosynthetic acclimation. Even so, it has again been illustrated in the CO$_2$ research that knowledge regarding individual leaf photosynthetic capacity provides little ability to predict plant growth (Körner 1996; Luo et al. 1997). Respiratory responses to elevated [CO$_2$] have been examined extensively, and a large amount of experimental data is available. Although some of the observed variability in respiratory responses to elevated [CO$_2$] can be explained conceptually, it seems imperative to evaluate various hypotheses in quantitative frameworks. The study of nutrient uptake kinetics is in its infancy, and some physiological processes reflect the availability of all the resources required for plant growth and thus become scaleable predictors of environmental conditions and resource availability. The validity of this convergence hypothesis has been made evident by the photosynthesis-nitrogen relationship at the leaf level, nitrogen-light at the canopy level, and light-use efficiency at the ecosystem scale. Although it varies in plot-scale measurements, Field (1991) suggested that light-use efficiency (defined as the ratio of dry matter production to the integrated energy absorbed) is a reasonable predictor of net primary productivity at large scales in combination with remote sensing data. In addition, Polglase & Wang (1992) and Luo & Mooney (1996) have recently examined leaf photosynthetic sensitivity to an increase in [CO$_2$] (normalized photosynthetic response to a small increase in [CO$_2$]) and found that it is independent of interspecific variation and growth environments for C3 plants and only varies with measurement temperature. This independence has the potential to simplify global extrapolation of leaf-level studies to provide a baseline prediction of a marginal increment in global carbon influx stimulated by a marginal increase in atmospheric [CO$_2$] (Luo et al. 1996). However, the actual increment in global carbon influx as stimulated by rising atmospheric [CO$_2$] has to account for aclimative changes in leaf, plant, canopy scales, temporal shifting of growing seasons, and biome movement (Luo & Mooney 1995).
The quest for predictive understanding
The quest for predictive understanding of plant responses to elevated \( \text{CO}_2 \) is in nature to identify general principles underlying diverse phenomena. A few historical examples may help illuminate this kind of endeavor. Gregor Mendel crossed flowers of Garden Peas and revealed the general principle of heredity. Brouwer (1962) developed the functional balance theory as a general concept to explain carbon partitioning. This was accomplished by pruning roots and cutting leaves of corn, rye, and bean plants together with manipulation of nutrient supply. Farquhar et al. (1980) developed a photosynthesis model by combined theoretical analysis of rubisco kinetics and synthesis of biochemical data. This model now provides transdisciplinary applications in plant physiology, ecosystem and global biogeochemical cycles.

It is a legitimate question for scientists in the plant \( \text{CO}_2 \) research area to ask: how can we develop general biological principles underlying variable plant responses to elevated \( \text{CO}_2 \)?

Simple experiments, like Mendel's work in genetics, are unlikely to reveal general principles in the plant-\( \text{CO}_2 \) research because \( \text{CO}_2 \) affects plants both directly and indirectly in a complex way. As discussed, rising atmospheric \( \text{CO}_2 \) has direct effects on photosynthesis and respiration. These effects can then be translated to various degrees into changes in whole plant growth, allocation, and morphology. The changes in turn have feedbacks to lower hierarchical levels causing, for example, changes in leaf photosynthetic capacity. Feedbacks on higher levels potentially alter community dynamics and ecosystem carbon and nutrient cycling. To develop general principles for such a complex system requires close collaboration between modelers and experimentalists. Towards this end, critical experiments guided by theoretical considerations, as Brouwer (1962) conducted on plant responses to nutrients, will be useful and effective. Combining theoretical and experimental studies of physiological mechanisms, as Farquhar et al. (1980) and others accomplished for photosynthesis, will become essential in developing our predictive understanding of plant responses to elevated \( \text{CO}_2 \).

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