Running head: Control and Adjustment of the Rate of Photosynthesis Above Present CO_2 Levels.

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A Characterization of the Regulation of Light-Saturated Photosynthesis As It Departs Rubisco Limitation at Above-ambient p(CO_2): What is Likely To Determine Photosynthetic Rates in the Future Atmosphere?

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Footnotes:

This work was supported by the Electric Power Research Institute (RP3041-02) and the United States Department of Energy Terrestrial Carbon Processes Program (DE-FG03-95ER62013/A000) and the Global Climate Change Program of the United States Department of Interior, Bureau of Reclamation, Cooperative Agreement No. 2-FC-30-00200, through the Lower Colorado River Region.

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Abstract

The adjustment of photosynthesis to different environmental conditions and especially to elevated CO_2 is often characterized in terms of changes in the processes that establish (limit) the net CO_2 assimilation rate. At slightly above present ambient pCO_2 light-saturated photosynthetic responses to CO_2 depart limitation by the catalytic capacity of tissue rubisco content. An hypothesis attributing this departure to limited thylakoid reaction/electron transport capacity is widely accepted, although we find no experimental evidence in the literature supporting this proposition.. The results of several tests point to the conclusion that the capacity of the thylakoid reactions cannot be generally responsible for the deviation from rubisco limitation. This conclusion leaves a significant gap in the interpretation of gas exchange responses to CO_2. Since the inputs to the photosynthetic carbon reduction cycle (CO_2 and photon-capture/electron-transport products) do not limit photosynthesis on the shoulder of the $A=f(c_i)$ curve, the control of photosynthesis can be characterized as: due to feedback. Several characteristics of gas exchange and fluorescence that occur when steady-states in this region are perturbed by changes in CO_2 or O_2 suggest significant regulation by conditions other than directly by substrate RuBP levels. A strong candidate to explain these responses is the triose-phosphate flux / inorganic phosphate regulatory sequence, although not all of the gas exchange characteristics expected with "TPU-limitation" are present (e.g. oxygen-insensitive photosynthesis). Interest in nitrogen allocation between rubisco and light capture / electron transport as the basis for photosynthetic adjustment to elevated CO_2 may need to be reconsidered as a result of these findings. Contributors to the feedback regulation of photosynthesis (which may include sucrose phosphate synthase and fructose bisphosphatase activities, phloem loading, and "sink-strength") may play a large role in the adjustment of photosynthesis to elevated CO_2. The continuing rise in atmospheric CO_2 elevates the need to understand the regulation of photosynthesis that is not related to rubisco capacity.

Introduction

Studies of photosynthetic gas exchange often use derivatives of the Farquhar model (e.g. Farquhar, Caemmerer, and Berry, 1980; Farquhar, and von Caemmerer, 1982; Harley, and Sharkey, 1991, Harley *etal.*1992) to quantify the three capacities that, in sequential ranges of CO_2, are considered to limit photosynthesis. Proceeding from limiting to rate-saturating levels of CO_2, these capacities are: (1) the quantity of the carboxylating enzyme, rubisco; (2) the maximum capacity for thylakoid reactions/electron transport; (3) the capacity for sucrose and starch synthesis as reflected by the release of inorganic phosphate, from triose-phosphates. Current high interest in plant responses to rising concentration of atmospheric CO_2 is one reason that studies now focus on limitations of photosynthesis other than rubisco. This report concerns the second of the capacities expected to be limiting — electron transport. The electron transport capacity is thought to limit photosynthesis when atmospheric CO_2 is in the range of roughly 40 to 60 Pa.

Specifically, the current common interpretation of measured net CO_2 assimilation as a function of leaf intercellular CO_2 pressure, $A=f(c_i)$ [abbreviations in Table 1], is as follows. When c_i is in the range of roughly 4-35 Pa, photosynthesis conforms to a rectangular hyperbolic relationship between CO_2 and demonstrable tissue rubisco content. In this region photosynthesis is considered rubisco-capacity limited (Sage, *et al.* 1990; Seemann, *et al.*, 1981; von Caemmerer and Farquhar, 1981). As c_i is increased above about 35 Pa, the $A=f(c_i)$ response diverges sharply, falling below the rubisco-dependent hyperbola. This divergence from rubisco-limitation is considered to result from less than saturating re-supply of rubisco's RuBP substrate at both limiting Photosynthetic Photon Flux Densities (PPFD) and saturating

PPFD (Farquhar, *et al.* 1980). At low PPFD, the capture of photons can obviously be limiting. At saturating PPFD the absorbtion of photons is not expected to be limiting and this expectation is confirmed by the fact that high energy (or non-photochemical) quenching of chlorophyll (q_e) generally remains high. At saturating PPFD the expected limitation in supply of RuBP, is ascribed to the photosynthetic rate having reached the maximum capacity for thylakoid-based reactions, J_{max} , (Farquhar *et al.* 1980; Farquhar and von Caemmerer, 1982; von Caemmerer and Farquhar,1981). The photosynthetic rate, J_{max} , and electron transport are related through the stoichiometeric requirement for RuBP regeneration. Empirically, photosynthesis continues to rise as a function of CO_2 in the region where J_{max} is considered limiting. In the context of the Farquhar model's structure of a sequence of single limitations this was explained (Farquhar and Caemmerer, 1982) as competitive diversion of the J_{max} dictated (fixed) quantity of RuBP toward rubisco's carboxylation (from oxygenation) reaction. As c_i is raised further, to perhaps 80 Pa, steady-state photosynthesis approaches CO_2 saturation and, at the same time, is often found to be unaffected by reductions in the rate of photorespiration (photorespiration being reduced as a result of low $p(O_2)$). This cannot be explained in the context of rubisco's carboxylation and oxygenation reactions competing for limiting RuBP (Sharkey 1985a,b). Sharkey therefore proposed the existence of the third limiting capacity, under which the rates of RuBP production and photosynthesis are controlled by the rate of inorganic phosphate release (in sucrose and starch synthesis) acting via a limited supply of P_i for ATP synthesis. While it is a central point of the Farquhar model framework that $A=f(c_i)$ points not on the initial, rubisco-capacity-defined hyperbola result from non-saturating levels of RuBP. Nonetheless, it is now accepted that in the steady-state at low PPFD the RuBP pool does not decline to an extent that explains the observed photosynthetic rate. Rather, the activation state of rubisco is modulated, holding the RuBP pool high, and, parenthetically, causes rubisco to retain more of the proximal control over photosynthesis than had been

expected (Perchorowicz *et al.*, 1981; Mott *et al.*, 1984; Woodrow and Berry, 1988). Some evidence exists of similar regulation occurring as c_i is varied but the degree to which rubisco is held near RuBP saturation remains uncertain (Sage 1990; Sage *et al.*, 1988,1990; Sharkey *et al.*, 1986; Woodrow and Berry, 1988). One of the several consequences of rubisco activation and the capacity for RuBP regeneration declining at the same time would be that the contributions of different potential sources of photosynthetic rate control could not be separated on the basis of the RuBP pool size nor the shape of the steady-state $A=f(c_i)$ alone.

Is there justification for raising the question of whether or not electron transport capacity is a significant source of limitation to photosynthesis? Physical correlations have been demonstrated between extractable rubisco activity *vis-à-vis* the initial slope of $A=f(c_i)$ curves (Seemann *et al.*, 1981; Woodrow and Berry, 1988). Limiting sucrose synthesis, restrictive levels of free P_i and O_2-insensitive photosynthesis have also been correlated (Sharkey, 1986b; Woodrow and Berry, 1988). The evidence offered by von Caemmerer and Farquhar (1981) in support of the existence of an electron transport limitation is a correlation between gas exchange measured *in vivo* and *in vitro* measurement of electron transport activity extracted from the same leaves. The dangers of loosing activity from *in vitro* measurements and potentially under-estimating capacity are well known. In the time since that report of limited electron transport capacity *in vivo* studies of the redox state of photosynthetic transport intermediates have provided more reliable information on electron transport limitations and rates. From such measurements Weis and co-workers (1986) found that, at saturating light, energy-dependent fluorescence quenching remains high and electron transport activity remains under the control of electron donation to the photosynthetic electron transport chain over the entire $A=f(c_i)$ curve. Following this, Weis and Berry (1987) showed that fluorescence quenching via electron transport is closely controlled in proportion to the demand for electron

transport products in the PCR cycle. Those findings suggest, but do not prove, the lack of a limitation due to electron transport capacity. This information notwithstanding, the view that, at saturating PPFD, the capacity for electron transport (J_{max}) causes the $A=f(c_i)$ relationship to deviate from the initial rubisco – capacity – related hyperbola has remained an important part of the general understanding of photosynthetic gas exchange. In this communication we report observations that caused us to question the involvement of limited electron transport capacity in photosynthetic CO_2 response curves. We then report the results of two tests of the hypothesis that this capacity causes photosynthesis to deviate from rubisco limitation.

Methods and Materials

Plant Material

Plants were grown in the following situations: (1) *Pinus ponderosa* Laws. in open-top chambers in Placerville, CA, at ambient $p(CO_2)$ in the high soil nitrogen treatment of an experiment described in (Ball *et al.*, 1992; Johnson and Ball, 1996); (2) *Populus Fremontii* Wats. seedlings in well watered and fertilized 44 liter pots in natural – light growth chambers in 33/15°C day/night temperature regime, (3) *Populus tremuloides* Michx. seedlings in 20 liter pots in these same growth chambers set to temperatures of 25/15 °C; (4) *Glycine max* L., *Nicotiana tabacum* L., and *Prosopis juliflora* (Sw.) DC. var *Torreyana* L. Benson. seedlings in well watered and fertilized 10 liter pots in a greenhouse in Reno, NV with temperature maxima and minima set to 28 and 12°C, respectively.

Measurements

Gas exchange measurements on *P. ponderosa*, *P. Fremontii*, *P. tremuloides*, *P. juliflora*, and *G. max* were made using open-flow gas exchange systems as described by Ball (1987). Other measurements on *G. max*, *P. Fremontti*, and *N. tabacum* employed a multi-channel rapid response gas exchange system described by Oja (1983). Photosystem II fluorescence was measured with a pulse – modulated – system configured as described in Daley *et al.*, (1989). Calibrations of fluorescence measurement for electron transport on *G. max*, and *P. Fremontii*, followed Weis and Berry (1986) and on *N. tabacum* followed Gentry *et al.*, (1989).

Parameters and Equations

Physiological parameters and relationships used here (or equations from which they are derived) can be found in Farquhar and Caemmerer (1982) or Woodrow and Berry (1988).

The rate of photosynthetic carbon fixation (μmol m^{-2} s^{-1}) is

$$P = v_C \ (1 - 0.5\ v_O) \qquad (1)$$

in which v_C and v_O are, respectively, the carboxylation and oxygenation rate of RuBP at rubisco. Gas exchange measures net CO_2 assimilation (A) which differs from P by the amount of normal respiration occurring in the light:

$$P = A - R_d. \qquad (2)$$

Estimates of R_d are from gas exchange measurements around the photo-compensation point, Γ^*, and/or by measurement of gas exchange on darkened leaves. The parameter Γ^* is the intercellular CO_2 pressure (c_i) at which v_O / v_C = 2 and is taken as

$$\Gamma^* = 0.5\ o_i \ / S \qquad (3),$$

where (o_i) is intercellular oxygen pressure and S is the rubisco specificity factor: 2360 Pa $[CO_2]$ Pa^{-1} $[O_2]$ **(14)**. From gas exchange measurements, the rate of RuBP carboxylation is

$$v_C = P / (1-(0.5\ o_i\ / S\ c_i)) \quad (4),$$

and the rate of RuBP oxygenation is,

$$v_O = (v_C\ o_i\ / S\ c_i) \quad (5).$$

Calculation of electron transport requirements to regenerate RuBP following carboxylation or oxygenation depend upon assumptions of how requirements for ATP and NADPH are balanced. While Farquhar and von Caemmerer (1982) prefer one set of values, they point out that the balance is not known and not fixed. Therefore, we use the values most biased against our arguments by taking the electron transport rate needed to support photosynthesis as,

$$ETR = 4\ v_C + 4\ v_O \quad (6).$$

Using other values would not materially influence this analysis. From this, the steady-state rate of RuBP production and consumption is ETR/4.

Farquhar *et al.* (1980) suggested that the catalytic capacity of rubisco in tissue, $V_C = [E] \cdot k_{cat}$, could be conveniently determined from dA/dc_i around Γ^* (their Eq. 42). The slope is used to eliminate the need to know R_d, and the vicinity of Γ^* was suggested to assure saturating RuBP. This approach has been widely used. However, a value for the quantity of active and RuBP-saturated rubisco required to support observed P can actually be estimated at any c_i:

$$V_C^R = v_C\ (c_i + K_C\ (1+o_i\ /K_O)/\ c_i\ ) = P\ (c_i + K_C\ (1+\ o_i\ /\ K_O)/(\ c_i\ - (0.5\ o_i\ /S)) \quad (7).$$

This parameter is less than V_C in approximate proportion to the product of the rubisco activation state and the degree of RuBP saturation — when activation and saturation are

expressed as decimal fractions (see Woodrow and Berry, 1988 Eq. 4). We find the highest values for V_C^R at or just prior to $P=f(c_i)$ departing the "rubisco-dependent" portion of the curve and use these highest values to define the rubisco – capacity – dependent hyperbola.

Results

When c_i is experimentally varied under saturating PPFD (Figure 1), the relationships between steady-state photosynthesis, photosystem II fluorescence, and electron transport can appear at odds with the hypothesis that the capacity for electron transport causes photosynthesis to diverge from the rubisco-related initial hyperbolic curve. In the responses shown the peak electron transport rates occur at values of c_i at or just beyond the point where photosynthesis departs the rubisco-dependent curve (Figure 1 Panels A&B, C&D, E&F). Then, as c_i is increased further, there is a clear tendency for electron transport (and RuBP consumption/regeneration) to decline even as photosynthesis continues to rise. Thus, if there is limited capacity for electron transport, that capacity is not always needed to support photosynthesis observed in this region. Interestingly, this ETR decrease contrasts with what seems to occur when photosynthesis departs from the initial curve at low PPFD (Figure 2). There, electron transport often continues to increase as electrons become limiting with increasing c_i.

In an attempt to determine if electron transport generally rises, stays constant, or declines after the breakpoint of $A=f(c_i)$ curves we have analyzed (using fluorescence and/or stoichiometery) more than 150 different samples of *Pinus ponderosa*, *Populus tremuloides*, *Populus Fremontii*, *Gossypium hirsutum*, *Helianthus annuus*, *Glycine max* and *Prosopis julifora* as well as curves

published in von Caemmerer and Edmondson, 1986; Sharkey *et al.*, 1988; Sage *et al.*, 1988; Sharkey 1985a; Laisk and Sumberg, 1994. From a total of 160 electron transport curves examined, 124 showed declines soon after the $A=f(c_i)$ curve departed rubisco dependency. A group of 36 curves, mostly from *P. ponderosa*, showed either a constant or slightly increasing electron transport rate over an interval of 25 Pa after departing the initial hyperbolic curve. Those plants in which electron transport declined after $A=f(c_i)$ departed the initial curve could not have been limited by the capacity for electron transport since they did not utilize all the capacity they had demonstrated at lower c_i. For plants where electron transport was constant or increased in this region of the CO_2 response curve, electron transport cannot be eliminate as a contributor to the limitation of photosynthesis, however other aspects of regulation might also be involved.

A stronger test of the hypothesis is needed and can be made relative to the following prediction: a manipulation that diverts the potentially limiting supply of electrons (actually RuBP) away from the carboxylation to the rubisco oxygenation reaction should cause photosynthesis to depart the initial curve at lower c_i but at the same ETR. This prediction can be examined by varying $p(O_2)$. We obtained $P=f(c_i)$ values for leaves of *Glycine max* centered around the breakpoint of the response curve at 20.9 kPa O_2 (Figure 3). We then set different oxygen pressures and measured photosynthesis over similar ranges of c_i. Raising $p(O_2)$ from 20.9 to 40 kPa caused several changes in the operation of photosynthesis. The ETR was higher and increased between each point in this range of c_i (Figure 3 Panel B). At the same time, no $P=f(c_i)$ points departed from the initial hyperbolic response (dotted lines defined from V_C = 106 μmol m^{-2} s^{-1} in Figure 3 Panel A). In comparison, lowering the O_2 pressure to 10.2 or 5 kPa raised the photosynthetic rate, lowered ETR, made the break in the $P=f(c_i)$ curve sharper, and caused a more pronounced decline in ETR at c_i above the break. In this experiment, curves

with lower electron demand but higher photosynthetic rates departed rubisco dependency at lower values of c_i. Conditions that raised the requirement for electron transport but lowered the rate of photosynthesis kept $P=f(c_i)$ on the initial hyperbolic curve. These responses are the opposite of the prediction above.

Another important approach to probing the capacity for electron transport is to examine transient responses: where regulation masks system capacities to a smaller extent. Figure 4 Panel A shows a steady-state $P=f(c_i)$ curve for *Nicotiana tabacum*. During the measurement of that curve a change in cuvette $p(CO_2)$ from 20 Pa (c_i = 13.3) to 70 Pa was imposed while gas exchange and fluorescence transients containing four distinct features (numbered arrows of Figure 4 Panel B) were recorded between the steady states. The dual stream gas exchange system (Oja, 1983) used for these measurements has a relaxation time of approximately 1s. Thus data from the first 3s after the imposed change are not included in the figure. The sharp peak and trough in CO_2 uptake (#1) lasting some 10s likely resulted from re-organizing the PCR/PCO pools. Photosynthesis then rose to 1.34 times the eventual new steady-state level (#2). To support this additional carbon fixation the calculated electron transport rate also rose — reaching 1.37 times the new steady-state value before declining. The decline (#3) to approximately the new steady-state had a relaxation time of approximately 225s. Following these larger changes are several smaller and longer-term adjustments (#4) toward steady-state. Photosystem II fluorescence measured simultaneously confirmed that electron flow to photochemical processes increased and relaxed with kinetics very similar to photosynthesis. To avoid disturbing the system during the transient, we chose not to measure pulse saturated fluorescence during the relaxation. Thus we cannot actually calculate ETR from fluorescence during that period. At the steady states before and after the transient, energy-dependent quenching coefficients were 0.788 and 0.675 for c_i= 13.3 and 39.8, respectively. The RuBP-

saturated rubisco capacity required (V_C^R) to yield the observed initial steady-state and peak (#2) values of photosynthesis were 26.8 and 27 μmol $m^{-2}s^{-1}$, while relaxation to the new steady-state brought V_C^R down to 21.2. The electron transport system met demands (as required by V_C^R and c_i) more than 30% above the steady-state requirement in this region where J_{max} has been thought to limit photosynthesis.

Discussion

From the above results the hypothesis: that, at saturating PPFD, limited capacity for thylakoid reactions causes the departure of the $P=f(c_i)$ relationships from the initial rubisco-capacity-dependent hyperbola, appears to be incorrect. The data show that in air of normal oxygen concentration, at levels of c_i above the CO_2 response curve breakpoint all electron transport capacity is not employed. Increasing PCO cycle activity by raising the surrounding O_2 pressures induced more electron transport in the steady-state showing that limited thylakoid reactions need not be invoked to explain the departure of $A=f(c_i)$ curves from their initial hyperbolic projection (Figure 3). In contrast, lower than normal O_2 conditions (which reduces PCO cycle flux) increased PCR cycle flux, reducing the demand for electron transport but resulting in photosynthesis that appears less dependent on rubisco capacity. The contrast between photosynthesis at different levels of $p(O_2)$ shown in Figure 3 is a well known pattern that can be found in many published reports (e.g. Laisk and Sumberg, 1994; Sharkey, 1985b; von Caemmerer and Farquhar, 1981). From the oxygen experiment it seems that departure from the initial hyperbola was more closely related to the rate of carboxylation (v_C) than any other photosynthetic parameter.

The gas exchange transient shown in Figure 4 constitutes evidence that regulation — which brings the PCR/PCO cycles and electron transport below their maximum capacities — is important in determining photosynthesis in the region of the $A=f(c_i)$ curve beyond the initial hyperbolic dependence on c_i. Transients with a shape inverse that in Figure 4 occur when one changes from high c_i to a level of c_i within the initial hyperbolic region of CO_2 response curves (data not shown). These kinetics are slow enough that virtually any open flow gas exchange system can document them. We have often encountered similar transient responses during gas exchange measurements but previously ignored them. The time constant for the transient between points 2 and 3 in Figure 4 is similar to time constants reported for changes in rubisco activation state (Mott *et al.*, 1984; Woodrow and Berry, 1988). Sage et al. (1988) show similar transient responses upon raising the ambient CO_2 to very high (160 Pa) levels. Those workers also demonstrated that the regulation that occurred involved deactivation of rubisco and maintenance of a relatively large RuBP pool. The oscillatory behavior of photosynthesis that can be induced above ambient $p(CO_2)$ levels (Leegood *et al.*,1985; Sage *et al.*, 1988) also demonstrates that greater capacities for electron transport and photosynthesis exist than are used in this region of steady-state $A=f(c_i)$ curves.

Conclusion

Our results lead to the conclusion that the capacity for electron transport cannot be considered uniformly responsible for PPFD-saturated photosynthesis breaking away from the initial hyperbolic response curve with increasing CO_2 pressure. There may be plants in which this capacity does enter into the definition of $A=f(c_i)$ curves, but we did not find clear evidence of this. This conclusion leaves a gap — between the initial $A=f(c_i)$ curve (related to rubisco-capacity) and "oxygen insensitive" photosynthesis (related to limited sucrose synthetic activity)

— in which we are unable to point to one particular limiting factor as explanatory the $A=f(c_i)$ response.

We noted in our introduction that data of von Caemmerer and Edmondson **(1986)**, Sharkey *et al.* **(1986)**, and Sage *et al.* **(1988,1990)** show that reduced rubisco activation/maintenance of relatively high RuBP pools occurs as CO_2 is varied in the $A=f(c_i)$ region beyond the initial hyperbola. We also noted that when regulated variables such as the RuBP pool size or the rubisco activation state receive input from more than one source the contributions to overall control of the individual sources are difficult to apportion on the basis of steady-state fluxes alone or pool sizes alone. It is even more difficult to apportion regulatory responsibility when the variables are tied together as are the RuBP pool and the rubisco activation state. Woodrow and Berry **(1988)** describe how a regulatory system operating through levels of PGA, DHAP, and P_i might coordinate the activities of multiple components of photosynthetic metabolism including rubisco activation state and the RuBP level. These authors also consider several reasons why carbon metabolism might be regulated in this way. The extent to which their hypothesized regulatory system explains $A=f(c_i)$ responses is largely untested, and the contributions of the several potential regulatory inputs need to be quantified. Woodrow and Mott **(1993)** developed a robust means of assessing the state of the photosynthetic system that combined gas exchange, metabolite pool size measurements, and modeling. From a limited set of measurements their model output suggested that the activity of the stromal-FBPase, cytosolic-FBPase and SBPase should be of increasing importance in controlling the rate of photosynthesis as the $A=f(c_i)$ curve departs the initial hyperbola. The fact that all carbon entering the PCO cycle may not return to RuBP (Harley and Sharky, 1991) could also emerge as an important contributor to rate control in this region of the $A=f(c_i)$ curve. Further investigation is needed.

We emphasize that none of the points that we have made invalidate predictions of photosynthetic rates *per se* from empirical fits of the Farquhar model. This includes rates in the thylakoid-/ J_{max} -limited region of $A=f(c_i)$ curves. The Farquhar model is a semi-empirical model and as such it can make accurate predictions even though the underlying mechanisms are not fully included or understood. For some purposes steady-state electron transport rates within the so called J_{max} -limited region are reasonably well approximated as constant. This study suggests that there may generally be reasons other than the capacity for electron transport underlying observations of near constant ETR.

While fully recognizing that "merely predicting" the rate of photosynthesis without regard to the underlying mechanism can be useful, we note considerable interest in predicting how both photosynthetic capacity and resource use in photosynthesis (particularly nitrogen and water) will adjust to growth in a wide ranges of environmental conditions. The current interest in plant responses to elevated atmospheric CO_2 pressure is only one example of this. If we have a clearer understanding of the mechanisms actually regulating the rate of photosynthesis at elevated CO_2, we will be better prepared to understanding and predict the adjustments in photosynthetic systems. When we examine changes in photosynthetic resource use under different environmental conditions, we are most likely to find consistent relationships between rates and resource levels if we can focus on the portion of the system that is actually determining the rate. We do not seem to have a solid understanding of photosynthetic rate limitation at CO_2 pressures much above present ambient levels. In this same vein, the fact that plants tend to operate at values of c_i close to the breakpoint of the $A=f(c_i)$ curve is a matter of significant ecological interest. That observation has often been cited as evidence that plants balance investment between CO_2 capture and photon capture (von Caemmerer and Farquhar,

1981; Wong *et al.*, 1985; Cowan, 1986). It seems quite likely that stomatal regulation keeping c_i near the CO_2 curve breakpoint does represent a balance in resource use. Yet investment in PCR cycle and sucrose synthetic pathway enzymes, the demand for the products of photosynthesis, and the availability of water may all be as much a part of the balance achieved as is investment in capacity for photon capture/electron transport and rubisco.

Acknowledgments: Peter Curtis and Rowan Sage asked JTB questions that stimulated this paper and provided important discussion. As he often does, Joe Berry gave the prefect hint on how to frame the problem. In this case the advise was, "Go back and read our rubisco review." We thank Yiqi Luo, Andrew Peterson, Linda Piehl, and Kevin Griffin for their comments as well.

Manuscript Received:

Manuscript Accepted:

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List of Figures

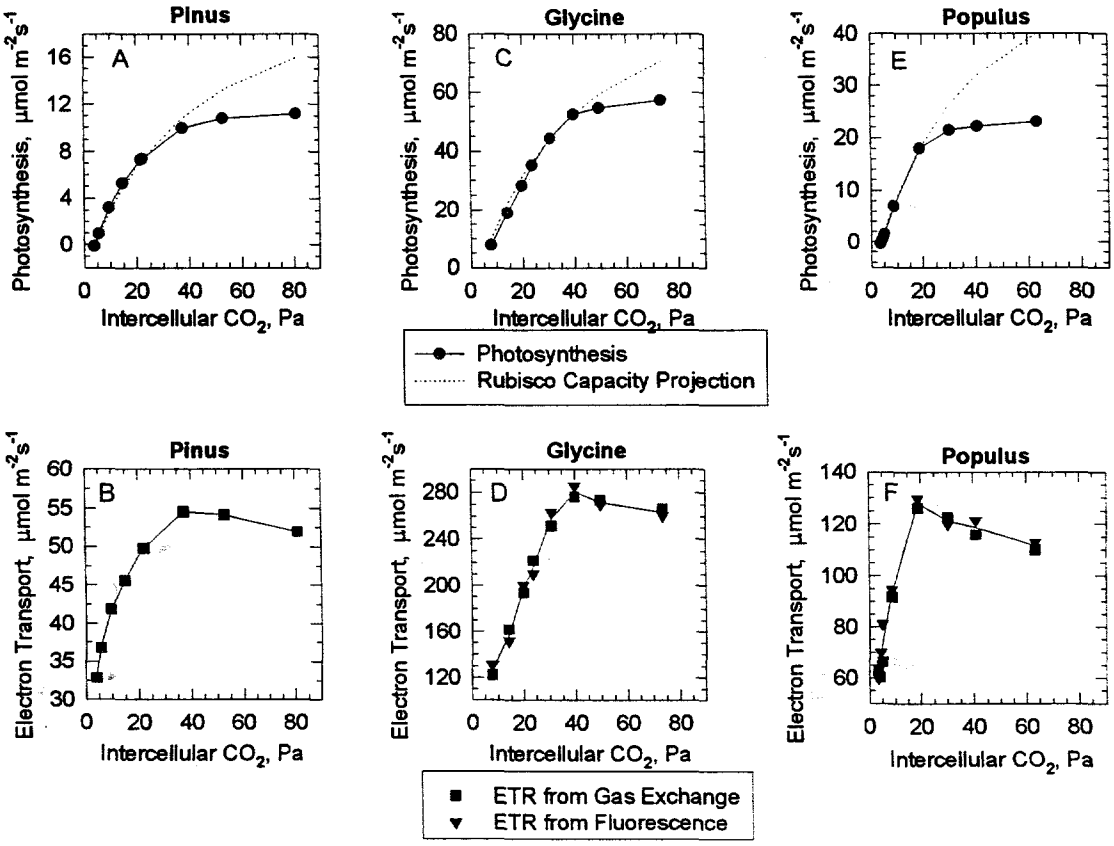
Figure. 1. Panel A, B, C The response of steady-state photosynthesis to variation in intercellular $p(CO_2)$ in, respectively, *Pinus ponderosa*, *Glycine max*, and *Populus tremuloides*. Panel D,E, F The electron transport rate for photosynthesis calculated from stoichiometery and confirmed by Photosystem II fluorescence through the CO_2 response curves shown in the respective panels above.

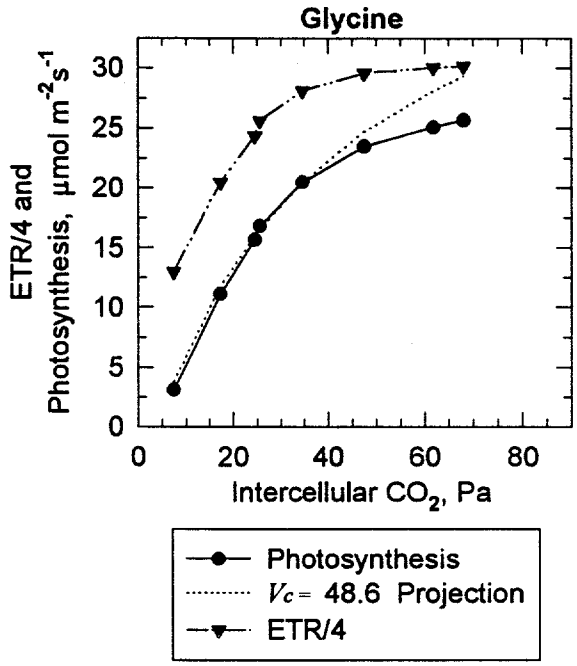
Figure 2. Photosynthesis and Electron Transport (the latter scaled by a factor of 4) as a function of leaf intercellular CO_2 pressure measure at PPFD of 500 μmol m^{-2} s^{-1} on a leaf of *Glycine max*. The graph shows that electron transport increases considerably, relative to Figure 1, after photosynthesis departs initial hyperbolic response to CO_2.

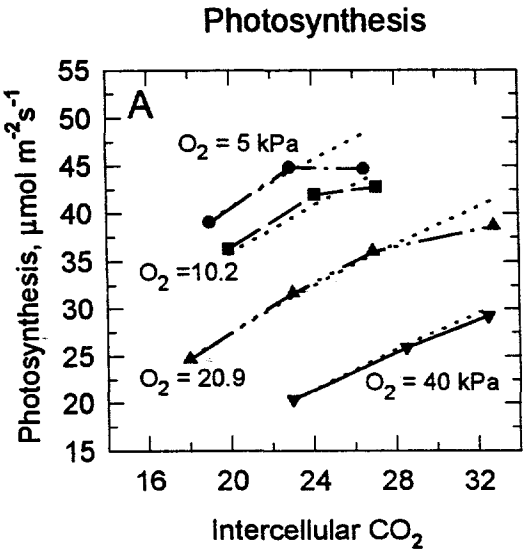
Figure. 3. Panel A Photosynthesis as a function of intercellular $p(CO_2)$ in *Glycine max* measured over the range of CO_2 covering the curve breakpoint at 5, 10.2, 20.9 kPa O_2 and over the same range of c_i at 40 kPa O_2. Panel B The calculated electron transport rates corresponding to the data in Panel A.

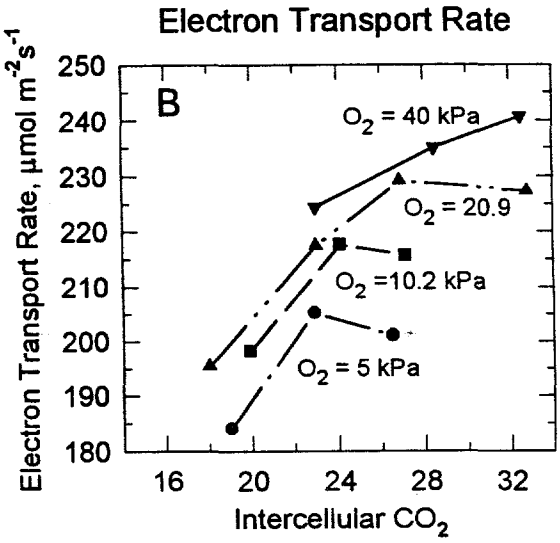
Figure. 4. Panel A. The steady state $P=f(c_i)$ curve for a leaf of *Nicotiana tabaccum* showing the transient movement of photosynthesis and electron transport when cuvette CO_2 was moved from 20 to 70 Pa. Panel B. The transient in photosynthesis and Photosystem II fluorescence produced by equilibrating a leaf of *Nicotiana tabaccum* at cuvette CO_2 of 20 Pa (c_i = 13.3 Pa) then switching ambient CO_2 to 70 Pa.

Table 1. List of Abbreviations and symbols used.

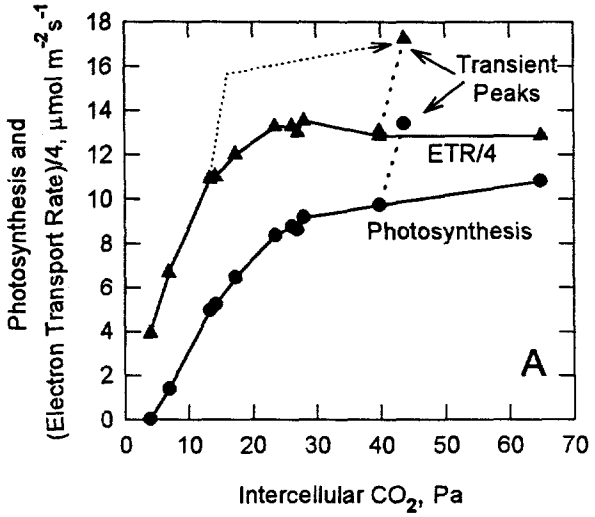

Pinus
A
Photosynthesis, µmol m-2s-1
Intercellular CO2, Pa
Glycine
C
Populus
E
Photosynthesis
Rubisco Capacity Projection
Pinus
B
Electron Transport, µmol m-2s-1
Glycine
D
Populus
F
ETR from Gas Exchange
ETR from Fluorescence


Glycine
ETR/4 and Photosynthesis, µmol m-2s-1
0
5
10
15
20
25
30
0
20
40
60
80
Intercellular CO2, Pa
Photosynthesis
Vc = 48.6 Projection
ETR/4


Photosynthesis
A
O_2 = 5 kPa
O_2 =10.2
O_2 = 20.9
O_2 = 40 kPa
Photosynthesis, µmol $m^{-2}s^{-1}$
Intercellular CO_2
55
50
45
40
35
30
25
20
15
16
20
24
28
32


Electron Transport Rate
B
O_2 = 40 kPa
O_2 = 20.9
O_2 =10.2 kPa
O_2 = 5 kPa
Electron Transport Rate, µmol $m^{-2}s^{-1}$
Intercellular CO_2
250
240
230
220
210
200
190
180
16
20
24
28
32

Nicotiana Steady-State Photosynthesis and Electron Transport with Peak Transient Values



Nicotiana transient from 20 to 70 Pa CO_2 at 20.9 kPa O_2

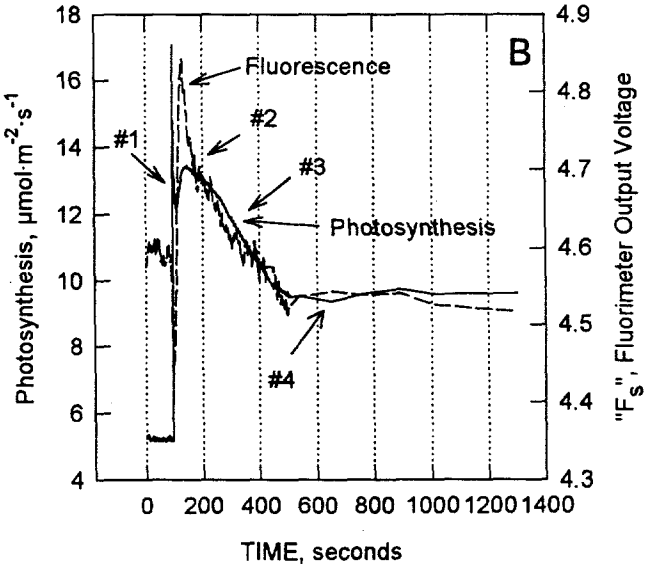


Table I. Abbreviations

A	Net CO_2 assimilation rate	μmol m^{-2} s^{-1}
Γ^*	Photo-compensation point	Pa
c_i	Leaf intercellular CO_2 pressure	Pa
DHAP	Dihydroxyacetone phosphate	
E	Holoenzyme (rubisco) concentration	
ETR	electron transport rate	μmol m2s
o_i	Leaf intercellular O_2 pressure	Pa
P	Photosynthetic rate	μmol m^{-2} s^{-1}
PGA	Phosphoglycerate	
R_d	normal Respiration rate	μmol m^{-2} s^{-1}
RuBP	Ribulose 1,5-bisphosphate	
S	rubisco specificity factor	Pa Pa^{-1}
v_C	velocity of RuBP carboxyation at rubisco	μmol m^{-2} s^{-1}
v_O	velocity of RuBP oxygenation at rubisco	μmol m^{-2} s^{-1}
V_C	Enzyme (rubisco) catalytic capacity	μmol m^{-2} s^{-1}
V_C^R	RuBP-saturated rubisco catalytic capacity required to yield observed P/c_i value	μmol m^{-2} s^{-1}