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Dear Editorial Board at *Nature*,

Atmosphere-biosphere carbon flux predictions diverge across terrestrial biosphere models when simulated using future climatic scenarios. Studies show that these models are particularly sensitive to the simulation of photosynthetic processes in response to increasing temperature and CO2 concentration1,2, yet few models incorporate explicit frameworks for simulating plant acclimation responses to such changes3.

Plants acclimate to increasing CO2 concentrations by reducing leaf nitrogen (N) allocation, stomatal conductance, and photosynthetic capacity, a pattern that often corresponds with acute increases in total leaf area and biomass accumulation at the whole plant level that dampen with time. N limitation has been hypothesized to be the primary mechanism driving increased growth rates under elevated CO2, as N availability commonly limits primary productivity and may decrease over time in elevated CO2 environments4–7. The N limitation hypothesis predicts that plants decrease leaf N allocation and photosynthetic capacity in response to progressive reductions in soil N availability due to elevated CO2. The N limitation hypothesis also predicts an acute stimulation in whole plant growth due to elevated CO2 that dampens over time as a result of progressive N limitation. However, this hypothesis does not explain why newly expanded leaves often have increased operational net photosynthesis rates, and limited empirical evidence exists supporting such an integrated role of soil nitrogen availability on leaf and whole plant responses to elevated CO27.

An alternative hypothesis to explain plant responses to elevated CO2 suggests that the reduction in leaf biochemical process rates and stomatal conductance is the result of an allocation strategy that allows plants to optimize resource use efficiency at the leaf level and maximize N allocation to whole plant growth. This hypothesis uses principles from optimal coordination theory to suggests that plants optimize leaf photosynthetic processes by optimally allocating N to Rubisco carboxylation such that net photosynthesis rates are equally co-limited by the maximum rates of Rubisco carboxylation and RuBP regeneration12,13. If true, the theory predicts that plants should respond to elevated CO2 by decreasing N investment to Rubisco, which increases operational net photosynthesis rates at reduced stomatal conductance rates while maximizing N allocation to structures that support whole plant growth. The expected optimal leaf response to elevated CO2 has recently received some empirical support14, but no studies to our knowledge have connected this leaf response to whole plant responses to elevated CO2. Importantly, this theory suggests that leaf responses to elevated CO2 should be independent of soil N availability, though does not discount the potential role of soil N availability on whole plant growth.

Finally, the method by which plants acquire nitrogen (“nitrogen acquisition strategy”) is likely an important factor when considering leaf and whole plant responses to elevated CO2, particularly because costs of acquiring nitrogen often vary in species with different acquisition strategies. Nitrogen acquisition strategy has been shown to regulate whole plant responses to elevated CO215,16. However, a better understanding regarding integrated leaf and whole plant mechanisms of such responses are needed, particularly if the strength of microbial symbioses change across resource availability gradients.

To disentangle effects of soil nitrogen availability and nitrogen acquisition strategy on leaf and whole plant responses to elevated CO2, we grew *Glycine max* (“soybean”) under one of two CO2 concentrations (420 ppm and 1000 ppm), one of two inoculation treatments (inoculated and uninoculated), and one of nine soil nitrogen fertilization treatments in a full-factorial growth chamber experiment. After seven weeks of development, we measured leaf nitrogen content, conducted net photosynthesis-by-intercellular CO2 concentration curves to estimate the maximum rates of Rubisco carboxylation and RuBP regeneration, and destructively harvested individuals total leaf area, and whole plant biomass. We also calculated the fraction of leaf nitrogen allocated to photosynthesis, structural carbon costs to acquire nitrogen, and the percent of leaf nitrogen acquired from the atmosphere.

In support of patterns expected from optimal coordination theory, elevated CO2 decreased the fraction of leaf nitrogen content allocated to photosynthetic tissue, the maximum rate of Rubisco carboxylation, and the maximum rate of RuBP regeneration. We also find that elevated CO2 decreases the maximum rate of Rubisco carboxylation more than the maximum rate of RuBP regeneration, allowing leaves to approach optimal co-limitation of Rubisco carboxylation and RuBP regeneration rates. In all cases, leaf responses to CO2 were independent of fertilization or inoculation treatment, suggesting that these responses were independent from nitrogen availability. Interestingly, we also find that elevated CO2 increased total leaf area and total biomass, responses that were enhanced with increasing fertilization and associated with reductions in the cost of acquiring nitrogen and resulting increases in nitrogen uptake.

Interestingly, our results provide support for both the progressive nitrogen limitation and optimal coordination hypotheses, suggesting that each hypothesis operates on a different scale. Specifically, increased whole plant growth under elevated CO2 was likely driven by an increase in plant nitrogen uptake, showing support for patterns expected from progressive nitrogen limitation. However, fertilization did not modify leaf responses to elevated CO2, and leaf physiological responses show strong evidence supportive of patterns expected from optimal coordination theory.

Elevated CO2 experiments rarely quantify both leaf and whole plant responses within the same experiment, and studies linking these responses often rely on meta-analyses from different studies. Our findings integrate both leaf and whole plant responses to elevated CO2 and connect these findings to changes in costs of acquiring nitrogen. Therefore, we feel this paper will reach a broad audience and will be cited amongst both the modeling and ecophysiological communities. As such, I submit this letter on behalf of my coauthors as pre-submission inquiry to *Nature* and the associated *Nature* family of journals to gauge interest in findings from this experiment.

Sincerely,

Evan A. Perkowski

*On behalf of coauthors Ezinwanne Ezekannagha and Nicholas G. Smith*