

RESEARCH PAPER

A meta-analysis of responses of canopy photosynthetic conversion efficiency to environmental factors reveals major causes of yield gap

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Received 7 February 2013; Revised 3 June 2013; Accepted 10 June 2013

Abstract

Improving plant energy conversion efficiency (ϵ_c) is crucial for increasing food and bioenergy crop production and yields. Using a meta-analysis, the effects of greenhouse gases, weather-related stresses projected to intensify due to climate change, and management practices including inputs, shading, and intercropping on ϵ_c were statistically quantified from 140 published studies to identify where improvements would have the largest impact on closing yield gaps. Variation in the response of ϵ_c to treatment type and dosage, plant characteristics, and growth conditions were also examined. Significant mean increases in ϵ_c were caused by elevated $[\text{CO}_2]$ (20%), shade (18%), and intercropping (15%). ϵ_c increased curvilinearly up to 55% with nitrogen additions whereas phosphorus application was most beneficial at low levels. Significant decreases in ϵ_c of -8.4% due to elevated $[\text{O}_3]$, -16.8% due to water stress, and -6.5% due to foliar damage were found. A non-significant decrease in ϵ_c of -17.3% was caused by temperature stress. These results identify the need to engineer greater stress tolerance and enhanced responses to positive factors such as $[\text{CO}_2]$ and nitrogen to improve average yields and yield potential. Optimizing management strategies will also enhance the benefits possible with intercropping, shade, and pest resilience. To determine optimal practices for ϵ_c improvement, further studies should be conducted in the field since several responses were exaggerated by non-field experimental conditions.

Key words: Climate change, crop management, harvest index, meta-analysis, photosynthesis, radiation use efficiency, yield potential, yield gap.

Introduction

Improving crop yields to meet the demands of the expanding population with increasing economic capacity is growing increasingly difficult. The human population is expected to reach almost 9 billion people by the middle of this century (Cohen, 2003). The rise in population coupled with improving living standards in China and India necessitate that yields double in less than 40 years (Tilman *et al.*, 2011), yet rates of yield increase in some major crops have slowed in the most recent

decades compared to rates of the 1970s and 1980s (Fischer and Edmeades, 2010; Long and Ort, 2010; Ray *et al.*, 2012). Meeting food and feed demands for the middle of the 21st century poses a formidable challenge to researchers and growers.

The Monteith equation (Monteith, 1977, as modified by Zhu, Long, and Ort, 2010) predicts that

$$Y = S_i \times 0.487 \times \epsilon_i \times \epsilon_c \times \epsilon_p$$

where yield potential (Y) is dependent upon the amount of incident solar radiation (S_t) received over the growing season, the proportion of S_t that is photosynthetically active radiation (PAR; estimated to be 0.487), the radiation interception efficiency of the plant (ϵ_i), the conversion efficiency of intercepted radiation into biomass (ϵ_c), and the partition efficiency of biomass into harvestable product (ϵ_p). The quantity of S_t during a growing season for a specific crop is largely predetermined by latitude, altitude, and time of year in a particular location, and the percentage that is PAR varies by location and can fluctuate within a day due to solar elevation and cloudiness (Monteith, 1965, 1972). Since the middle of the 20th century, ϵ_i and ϵ_p have been vastly improved due to management and breeding practices introduced during the Green Revolution, thereby closing the gap between actual and theoretical values (Evans, 1993; Hay, 1995; Sinclair, 1998). The greatest opportunity for yield potential improvement therefore lies in increasing ϵ_c (Zhu *et al.*, 2010).

Energy conversion efficiency (commonly referred to as 'radiation use efficiency' or RUE) is defined as the efficiency with which intercepted or absorbed energy is converted into biomass and is based on the photochemical efficiency of the entire plant canopy. Theoretical ϵ_c maxima have been calculated and show greater maximum efficiency in C_4 compared to C_3 plants; however, the greatest season-long ϵ_c values are estimated to reach only one-third to one-half of the theoretical maxima in common crops, making it the best candidate for increasing yield potential (Beadle and Long, 1985; Amthor, 2010; Zhu *et al.*, 2010). Raising yield potential is necessary to prevent stagnation in yield increase since the rate of yield increase will fall as average yields approach the existing yield potential. This widens the yield gap, or the difference between average and potential yields (Lobell *et al.*, 2009). However, the gap also increases, albeit detrimentally, when average yields decline or cannot keep pace with the yield potential increases of a given area and crop (Lobell *et al.*, 2009).

Major greenhouse gas concentrations, weather-related conditions associated with climate change, and management techniques including inputs, shading, and intercropping influence the realized yield potential in plants and perforce are among the most important causes of yield gap. Atmospheric CO_2 concentrations ($[CO_2]$) are predicted to reach 550 ppm by 2050, and background tropospheric ozone concentrations $[O_3]$ are projected to increase by 25% during the next few decades (Meehl *et al.*, 2007). Incidents of drought are predicted to become more frequent and extreme along with a 2–6 °C rise in terrestrial mean surface air temperature by the end of this century (Meehl *et al.*, 2007). While elevated $[CO_2]$ increases leaf photosynthesis of C_3 crops (Curtis, 1996; Curtis and Wang, 1998; Medlyn *et al.*, 1999; Wand *et al.*, 1999; Ainsworth *et al.*, 2002; Ainsworth and Rogers, 2007; Ainsworth, 2008), negative effects on leaf photosynthesis have been demonstrated by elevated $[O_3]$ (Morgan *et al.*, 2003; Wittig *et al.*, 2007; Ainsworth, 2008; Feng *et al.*, 2008), drought (Lawlor and Cornic, 2002), and extreme temperatures (Berry and Bjorkman, 1980; Oquist, 1983; Sharkey, 2005; Ruiz-Vera *et al.*, 2013). Reviews demonstrate a positive correlation between leaf photosynthesis and ϵ_c and suggest

that the above-mentioned climate/weather factors alter ϵ_c greatly when they affect leaf photosynthesis (Beadle and Long, 1985; Sinclair and Muchow, 1999). Leaf nutrient status also affects leaf photosynthetic rates, and therefore nutrient deficiencies, such as limited nitrogen and phosphorus, lower ϵ_c (Monteith, 1977; Beadle and Long, 1985; Sinclair and Muchow, 1999) and increase yield gap. Foliar damage from pests and pathogens has been shown to downregulate genes involved in photosynthesis (Bilgin *et al.*, 2010), suggesting an additional limiting factor to ϵ_c . When grown in the field, leaves at the tops of crop canopies often absorb light far in excess of what is needed for maximal photosynthesis, and the excess light must be dissipated as heat through non-photochemical quenching or as chlorophyll fluorescence to prevent damage to the photosynthetic apparatus (Ort, 2001). Reduction in incident radiation by shading decreases the amount of absorbed light energy at the top of the canopy, thereby increasing the efficiency with which absorbed light is used (Long *et al.*, 2006). Clouds, over topping leaves, as well as some synthetic shading materials increase the proportion of diffuse to direct radiation (Healey and Rickert, 1998) and therefore the total amount of light reaching shaded leaves within the canopy (Sinclair and Muchow, 1999), which, along with other factors, may be beneficial in intercropping (Anil *et al.*, 1998).

A plethora of literature exists on ϵ_c and how it is affected by climate change driving greenhouse gases, weather-related stresses predicted to worsen due to climate change, and management in the form of inputs and spatial planting arrangements. Therefore, a meta-analysis is a useful tool to statistically quantify the effects of these factors and thus quantify their importance in limiting yield. Previous reviews have described the results of ϵ_c studies (Monteith, 1977; Beadle and Long, 1985; Sinclair and Muchow, 1999), but none have quantified the effects of climate change, both causes and effects, and management on ϵ_c . The purpose of this study was to use a meta-analysis to (a) determine the percentage change in ϵ_c due to elevated $[CO_2]$, elevated $[O_3]$, water stress, temperature stress, nitrogen application, phosphorus application, foliar damage, shade, and intercropping, and (b) identify the effect of within treatment variability due to treatment type and dosage, plant characteristics, and growth environment on ϵ_c . Significant increases in ϵ_c were found with elevated $[CO_2]$, nutrient inputs, shading, and intercropping, and therefore these variables have the potential to raise the ceiling on yield potential, especially with further optimization and engineering for enhanced responses. Elevated $[O_3]$, water stress, temperature stress, and foliar damage decreased ϵ_c and warrant improved stress tolerance and management practices to prevent average yield decreases and ultimately a wider yield gap.

Materials and methods

Database development

Studies on ϵ_c or 'radiation use efficiency' were found using the Web of Science citation database (ISI, Philadelphia, PA, USA). Additional studies were included when referenced by the studies found by the methods above. Means, sample sizes, and standard

deviations (if available) were collected for both controls and treatments from individual studies (Supplementary Table S1, available at *JXB* online). In cases where other variances were given, standard deviations were back calculated, and if standard deviations were not reported, they were sought from the authors. When values were only available in figures, the values were digitized using Grafula 3 version 2.10 (Wesik SoftHaus, St Petersburg, Russian Federation). Values taken from the same study were considered independent when cultivars, location, growing period, stresses, and treatments or treatment levels differed (Ainsworth *et al.*, 2002).

The pool of available studies was narrowed by certain criteria. Studies that did not report sample size could not be used. ϵ_c values based on incident light were not included because apparent changes in ϵ_c may have been influenced by changes in ϵ_i (Monteith, 1994). Several studies included ϵ_c values during different parts of the growing season. When a value for the entire season was not available, a maximum value was used from vegetative stages since ϵ_c values determined from regression slopes of dry matter accumulation are more linear during vegetative stages (Sinclair and Muchow, 1999). If values based on both aboveground and total dry matter were included, only the value based on aboveground dry matter was used since methods of estimating or measuring belowground biomass varied greatly across studies. Values based on regression slopes of dry biomass versus intercepted radiation were used preferentially over calculated ratios due to lower errors (Monteith, 1994; Sinclair and Muchow, 1999). Comparisons between treatment and control were considered valid only if made within the same growing period and with equal growth conditions to avoid potential confounding effects. Although measurements and units of ϵ_c varied by study, these inconsistencies were circumvented using response ratios.

Meta-analyses

Traditionally, meta-analyses measure the response of several variables to a treatment. This study used a different approach in that it compared the change in one variable in response to several separate treatments. The response variable, ϵ_c , was compared across the atmospheric conditions of elevated [CO₂] compared to ambient conditions and elevated [O₃] compared to ambient air when in free air concentration enrichment (FACE) and charcoal-filtered air when in open-top chamber (OTC) studies. ϵ_c responses were also analysed in conditions predicted to occur due to climate change such as water and temperature stress compared to optimal water supply and temperature. The effects of management practices were examined using the treatments of nitrogen and phosphorus addition, foliar damage due to pests and disease, shading, and intercropping. For nutrient treatments, the control was the lowest amount of nutrient applied in a study, and treatments consisted of nutrient additions. In the foliar-damage treatment, the controls were healthy unaffected plants versus those that had suffered from pests or diseases, and shading analyses compared shaded plants to those in full sunlight. For the intercrop treatment, the control was a crop that was already established or the

crop that is more widely planted in the area and time period of the study (FAO, 2011), and the treatment occurred when another species was intercropped with the control species.

Categorical variables were used to test for heterogeneity within treatment groups. These consisted of treatment dosage or type (Table 1), plant characteristics, and growth condition (Table 2). Discrete variables were separated into their qualitative categories while continuous variables were assigned arbitrarily to categories.

The meta-analysis was conducted using the natural log of the response ratio (treatment mean/control mean; Hedges *et al.*, 1999; Rosenberg *et al.*, 2000). Values were reported as percentage changes where positive change represented stimulation and negative change represented a decrease in ϵ_c due to the treatment. A mixed-model analysis was run using MetaWin (Rosenberg *et al.*, 2000). When a sufficient number of studies contained standard deviation values within a treatment, a weighted parametric analysis was used (Gurevitch and Hedges, 1999) where the weight of a study was determined by its mixed-model variance (Hedges *et al.*, 1999). To test for ample standard deviations, both weighted and non-weighted analyses were run and the percentage changes were compared. Small changes (<10% difference between weighted and non-weighted) allowed for reporting weighted analyses whereas large changes or results that altered interpretations constituted reporting the results of the non-weighted, non-parametric analyses. Confidence intervals were determined by bootstrapping methods using 9999 iterations in non-parametric analyses (Adams *et al.*, 1997; Gurevitch and Hedges, 1999; Rosenberg *et al.*, 2000). In both weighted and non-weighted analyses, a treatment effect was considered significant if the 95% confidence intervals did not overlap zero. In non-weighted categorical analyses, categories within a treatment were considered significantly different if their confidence intervals did not overlap. The between-group variability (Q_b) of categorical variables in weighted analyses, determined according to Curtis and Wang (1998), was statistically tested using an alpha of 0.05.

Results and discussion

Atmospheric gas concentrations significantly affected the response of ϵ_c

Elevated [CO₂] increased ϵ_c by 20% overall (Fig. 1), which was similar to a reported 17% mean increase in yield due to elevated [CO₂] in FACE studies (Ainsworth and Long, 2005). The mean stimulation of ϵ_c was 2-fold greater for [CO₂] enrichment >599 ppm compared to enrichments of 450–599 ppm (Fig. 1). ϵ_c stimulation was significantly affected by photosynthetic type with a 5-fold greater stimulation in C₃ compared to C₄ plants (Fig. 1). This agreed with previous meta-analyses with a greater response in C₃ versus C₄ plants

Table 1. Categories and levels describing treatment dosage and type effects

Variable	Level 1	Level 2	Level 3	Level 4	Level 5	Level 6
[CO ₂] dosage (ppm)	450–599	>599				
[O ₃] dosage (ppb)	30–59	60–79	>79			
Water stress: type	Excessive	Limiting				
Water stress: timing	Vegetative	Reproductive	Growing season			
Temperature stress	Above optimal	Below optimal				
Nitrogen added (kg ha ⁻¹)	0–50	51–100	101–150	151–200	201–250	>250
Phosphorus added (kg ha ⁻¹)	0–50	51–100	>100			
Damage type	Pest	Disease				
Shade dosage (%)	0–50	51–100				

Table 2. Categories and levels describing plant characteristics and growth environment

Category	Level 1	Level 2	Level 3	Level 4	Level 5	Level 6	Level 7
Photosynthesis type	C ₃	C ₄					
Life cycle	Annual	Perennial					
Plant type	Crop	HerbNC	Tree				
Nitrogen fixation	Fixer	Non-fixer					
Growth condition	FACE	OTC	OTC (pots)	Field	Field (pots)	Greenhouse	Shelter (pots)

FACE, free air carbon enrichment; HerbNC, herbaceous non-crop; OTC, open-top chamber.

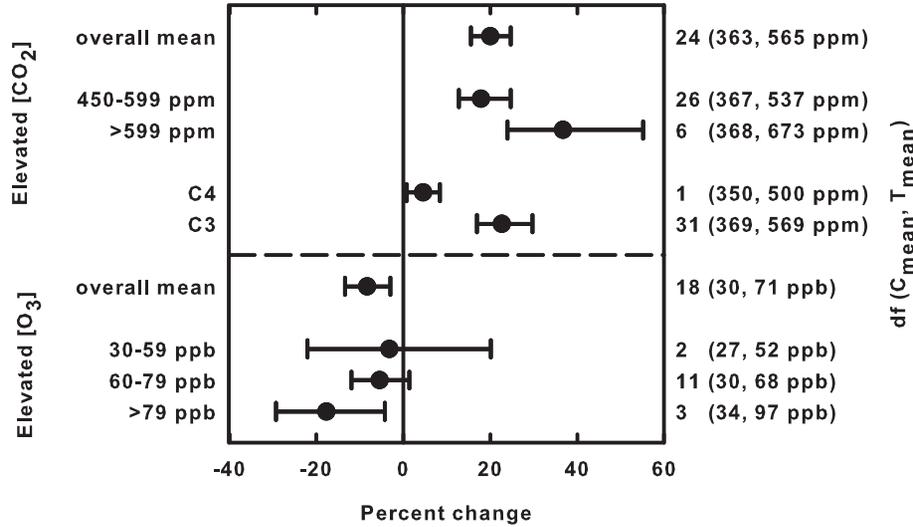


Fig. 1. Percentage change in ϵ_c due to treatments and categories within treatments in response to elevated [CO₂] and [O₃]. Symbols represent the mean percentage change from control conditions with 95% confidence intervals. Degrees of freedom (df) are listed on the right with mean control (C_{mean}) and treatment (T_{mean}) dosage in parentheses. Treatments are separated by dashed lines.

to elevated [CO₂] in variables related to ϵ_c such as A_{sat} and biomass accumulation (Wand *et al.*, 1999; Ainsworth and Long, 2005). However, the small sample size prevents strong conclusions in this area. Based on these findings, yield potential will increase with increasing [CO₂], which will beneficially widen the yield gap to allow for greater rates of increase in average yields. Engineering higher capacity forms of Rubisco, greater electron transport capacity, and stronger sink capacity will further enhance the responsiveness to elevated [CO₂] (Ainsworth *et al.*, 2008) and therefore overall yield potential.

Interactions with other variables may prevent realized yields from increasing with increasing [CO₂], resulting in a detrimentally wider gap between actual and potential yields. The response ratio was greater under elevated [CO₂] in wheat when background [O₃] was also elevated, but overall values of ϵ_c declined (Rudorff *et al.*, 1996). A 3-fold greater response ratio of ϵ_c to elevated [CO₂] was measured in a water-stressed legume compared to the well-watered condition, but again, the overall ϵ_c values were depressed by the negative stress (Clifford *et al.*, 1993). Elevated [CO₂] decreases stomatal conductance (g_s), which ameliorates the negative effects of other stresses (Leakey *et al.*, 2009) and increases ϵ_c to a greater extent under stressful conditions, but any increases in the magnitude of ϵ_c are absent in the presence of these stresses. Limited nitrogen often constrains the response of plants to

elevated [CO₂] (Stitt and Krapp, 1999); however, the only study that examined this interaction and its effects on ϵ_c did not report a change in the response or magnitude of ϵ_c in barley to elevated [CO₂] under limited nitrogen conditions (Manderscheid *et al.*, 2009).

Elevated tropospheric [O₃] significantly decreased ϵ_c compared to control conditions (-8.4%; Fig. 1), but the magnitude of the effect was considerably less than biomass and yield changes in wheat (Feng *et al.*, 2008), rice (Ainsworth, 2008), and soybean (Morgan *et al.*, 2003) exposed to elevated [O₃]. This suggests that O₃ also affects ϵ_p but the smaller impact on ϵ_c than on biomass is difficult to understand since any differences in ϵ_i are controlled for by using absorbed radiation in the ϵ_c calculation. The fact that studies that used ambient air as a control were included, suggest that these results may be more indicative of how increases in [O₃] will affect ϵ_c from present tropospheric concentrations rather than the level of inhibition caused by elevated [O₃] in relation to preindustrial levels and could explain the smaller biomass response in this analysis. Although the dose effect was not significant ($Q_b = 7.26$, $P = 0.08$), there was a trend for a greater decrease as the magnitude of elevated [O₃] increased (Fig. 1), which is consistent with past studies (Morgan *et al.*, 2003; Feng *et al.*, 2008). However, the lack of significance in this study may be due to the reasons postulated above. Photosynthesis

type and the type of life cycle did not affect the response of ϵ_c to elevated $[O_3]$ ($Q_b = 0.16$, $P = 0.64$ for photosynthesis type and $Q_b = 0.05$, $P = 0.84$ for life cycle), but again the C_4 category lacked sufficient degrees of freedom to draw solid conclusions (Supplementary Table S2). Contrary to these categorical results, previous reviews have emphasized that crop responses to elevated $[O_3]$ vary across and within species (Fiscus *et al.*, 2005) and have suggested there is potential for improvement using breeding and biotechnology to increase antioxidant capacity and improve $[O_3]$ sensing and signaling (Ainsworth *et al.*, 2008).

ϵ_c responded negatively to water and temperature stress

ϵ_c was significantly decreased by water stress (−16.8%; Fig. 2). The extent of the negative effects of water stress were significantly greater when water was limiting compared to when water was excessive (Fig. 2). The extent of water stress was not reported in a majority of the studies and therefore a relationship could not be determined between severity and changes in ϵ_c . The negative response of ϵ_c was greater when water stress occurred during reproductive stages or over the entire growing season compared to only during vegetative stages (Fig. 2), which would be expected to amplify its effect on yield for grain or fruit crops. In wheat, additional sinks during reproductive growth increase the demand for photosynthate resulting in greater leaf photosynthesis (Reynolds *et al.*, 2005), but water stress may prevent that photosynthetic stimulation and therefore cause a more pronounced decrease in ϵ_c as compared to the decrease evident during vegetative stages. Nitrogen-fixing plant responses to water stress were 3-times more negative than in non-fixing plants (Fig. 2). This finding portends concern regarding predictions of changing precipitation patterns considering legumes are the second most important plant

group to humans (Graham and Vance, 2003). An estimated 12–15% of the world's arable land is used for legume production and provides considerable benefits to both human vegetable protein consumption in addition to soil improvement through nitrogen fixation where fertilizers are limited (Graham and Vance, 2003). The 18% greater decrease in ϵ_c most likely contributes to reduced yields in legumes and subsequently planted crops on the same land, and it emphasizes the importance of improving drought tolerance and water use efficiency in legumes to increase ϵ_c under reduced precipitation. Although evidence was limited, elevated $[CO_2]$ may help mitigate the negative effects of drought in a legume crop since elevated $[CO_2]$ decreased the negative effects of drought in *Arachis hypogaea* by one third (Clifford *et al.*, 1993).

Temperature stress was the only treatment that resulted in a non-significant effect on ϵ_c (−17.3%) as compared to control growth conditions (Fig. 2), most likely due to the limited numbers of studies available. ϵ_c was not significantly affected by the type of temperature stress (Fig. 2), photosynthesis type, or nitrogen fixation (Supplementary Table S2), but it should be noted that photosynthesis type and nitrogen fixation categories were completely confounded with each other. One would expect temperature stress to cause a greater decrease in the ϵ_c of C_3 plants with above-optimal temperatures due to increased photorespiration, whereas ϵ_c would decrease to a greater extent in C_4 with below-optimal temperatures (Long *et al.*, 2006). However, the limited number of studies makes it difficult to make any conclusive remarks regarding the effects of temperature on ϵ_c as a function of photosynthetic type.

ϵ_c responses were greater with larger additions of nitrogen and smaller additions of phosphorus

The greatest changes in ϵ_c were evident with nitrogen applications. For the purpose of this analysis, nutrient additions

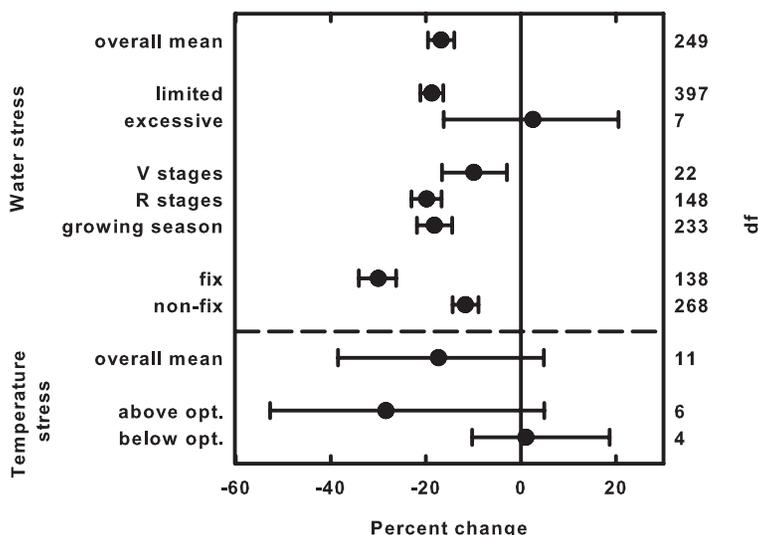


Fig. 2. Percentage change in ϵ_c due to treatments and categories within treatments in response to water and temperature stress. Symbols represent the mean percentage change from control conditions with 95% confidence intervals. Degrees of freedom (df) are listed on the right. Treatments are separated by dashed lines. V = vegetative; R = reproductive; fix = nitrogen-fixing plant; non-fix = non-nitrogen-fixing plant.

were used instead of leaf nutrient concentration due to the limited number of studies that reported leaf nutrient status. Since nutrient additions encompassed a large range (16–420 kg nitrogen and 17.5–111 kg phosphorus ha⁻¹), ϵ_c dosage responses were deemed more appropriate than overall responses. Increases in ϵ_c displayed a curvilinear response to nitrogen additions with a maximal response to nitrogen addition of about 55% occurring between 201 and 250 kg nitrogen added ha⁻¹ (Fig. 3). Regression analyses demonstrated a similar curvilinear trend with saturation in the ϵ_c response occurring after 250 kg nitrogen added ha⁻¹ (Supplementary Fig. S1). A similar curvilinear relationship exists between leaf nitrogen and leaf and canopy photosynthesis (Sinclair and Muchow, 1999). Approaching this from the opposite direction, crops could experience a 55% decrease in ϵ_c when nitrogen is limiting which would contribute significantly to yield gap. In many areas of the world where fertilizer is not available or affordable and soils are nutrient poor, it is very possible that decreases occur to this extent in ϵ_c , which could be translating to proportionally smaller yields under nitrogen limitation. Photosynthesis type and plant type significantly affected the response of ϵ_c to nitrogen. The stimulation in ϵ_c with added nitrogen in C₄ plants was 2.5-times greater than in C₃ plants (Fig. 3). The stimulation in herbaceous non-crops was almost 2-times greater than the response seen in crops, which may be due to growth in poorer or less intensively managed soils than crops. However, if genetic differences exist in nitrogen responsiveness, these could be used to make crops more responsive to nitrogen. Trees did not statistically differ from either crops or herbaceous non-crops (Fig. 3). Life cycle

did not significantly affect the response of ϵ_c to nitrogen additions ($Q_b=0.78, P = 0.59$; Supplementary Table S2).

Maximum increases in ϵ_c (36.3%) occurred with phosphorus additions of 51–100 kg ha⁻¹, but as application rates exceeded that dosage, the additional response in ϵ_c was drastically reduced (Fig. 3). However, only two studies contained phosphorus treatments greater than 100 kg ha⁻¹, and their results were quite variable. One demonstrated a large positive response of ϵ_c to phosphorus addition (Manrique, 1993) in contrast to the other study which reported a range of -4 to +5% percentage change in ϵ_c with additions of 111 kg ha⁻¹ (Plenet *et al.*, 2000). The phosphorus contents of the soils prior to fertilization in the treatment plots differed across studies and may have caused the disparity seen between studies. In Plenet *et al.* (2000), available soil phosphorus in all treatment levels prior to fertilization were considerably higher than those reported, both pre- and post-fertilization, by Manrique (1993). While available soil phosphorus concentration may have been a better metric to examine the effects of phosphorus additions on ϵ_c , the frequency with which this was reported was low. However, these findings suggest fine-tuning inputs based on soil phosphorus availability to maximize ϵ_c and minimize wasted phosphorus. Non-nitrogen-fixing plants demonstrated a significantly greater increase in ϵ_c (35.0%) compared to fixing plants (7.5%) when phosphorus was more readily available (Fig. 3), which emphasizes the need for sufficient soil phosphorus to prevent yield losses in legumes. Photosynthesis type did not significantly affect the response of ϵ_c to phosphorus (Supplementary Table S2).

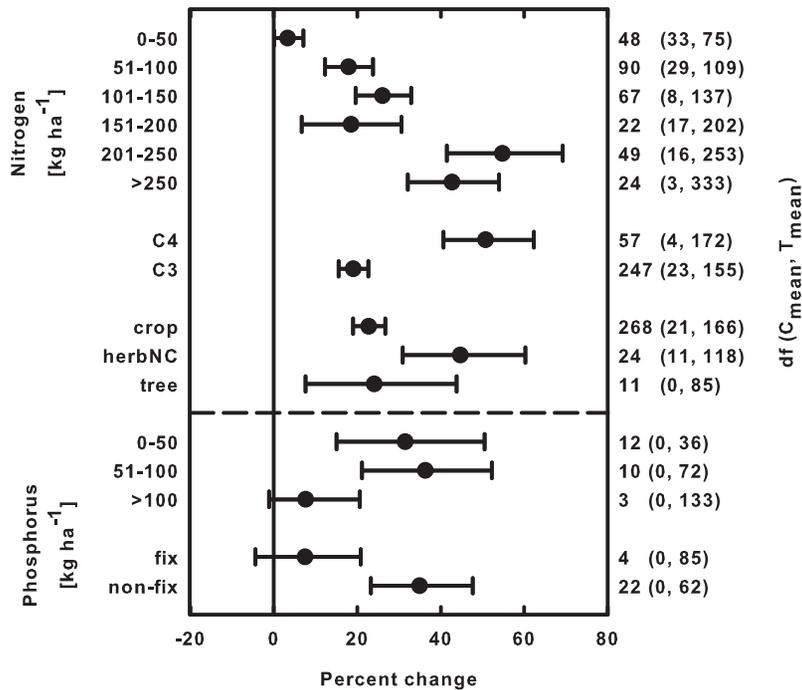


Fig. 3. Percentage change in ϵ_c due to treatments and categories within treatments in response to nitrogen and phosphorus additions. Symbols represent the mean percentage change from control conditions with 95% confidence intervals. Degrees of freedom (df) are listed on the right with mean control (C_{mean}) and treatment (T_{mean}) dosage (kg ha⁻¹) in parentheses. Treatments are separated by dashed lines. herbNC = herbaceous non-crop species; fix = nitrogen-fixing plant; non-fix = non-nitrogen-fixing plant.

Foliar damage significantly decreased ϵ_c whereas shading and intercropping significantly increased ϵ_c

Foliar damage demonstrated a relatively small but significant influence on ϵ_c (-6.46%; Fig. 4), and pest damage had greater effects on ϵ_c compared to damage from disease (-19.5 and -2.14 %, respectively; Fig. 4). The majority of data points representing pest damage came from one study on the effects of spider mites on ϵ_c (Sadras and Wilson, 1997). Percentage changes in ϵ_c ranged from -56% to -0.3%. However, these were correlated with greater mite populations, which also varied within the study (Sadras and Wilson, 1997) and may account for the greater disparity between pest and disease damage while also demonstrating the benefits of effective pest management to maximize ϵ_c . Perennials displayed a significantly greater decrease in ϵ_c response compared to annuals when damage occurred (Fig. 4).

Overall, shading significantly increased the response ratio of ϵ_c compared to plants in full sunlight by 18.0% (Fig. 4). Although the extent of shading did not result in a statistically significant difference in the response (Fig. 4), the data showed the expected trend of the greatest percentage increases in ϵ_c occurring at the lowest levels of shading. The potential for extraordinarily great changes in ϵ_c with shading (>200%

increase) were demonstrated in two studies with <50% shade treatments (Stirling et al., 1990; Cruz, 1996). Large increases in ϵ_c with shade were possible primarily because all measurements included in the shade analyses were conducted in the field in which ambient light levels are often well above saturating light levels needed to achieve maximum photosynthesis for leaves at the top of the canopy (Ort, 2001). Therefore, plants absorb several-fold more light than they can use photosynthetically, which, by definition, drastically reduces the photosynthetic efficiency of the leaf (Long et al., 1994). Shade not only decreases the amount of light reaching the canopy, but it can also change light qualities by increasing the ratio of diffuse to direct sunlight and allowing greater light penetration into the canopy to stimulate canopy photosynthesis (Roderick et al., 2001). While the large benefits of shade on ϵ_c could translate to greater yields, the fact that shade decreases overall S_t on a crop may negate those benefits on yield potential if excess light is inactively absorbed. However, using taller plants to limit radiation reaching light-sensitive canopies or optimizing light distribution in canopies through modified canopy architecture (Long et al., 2006) or chlorophyll content (Ort et al., 2011) could abate this problem. This analysis could not identify varying light sensitivity across plant categories since several plant characteristics within the shade treatment were

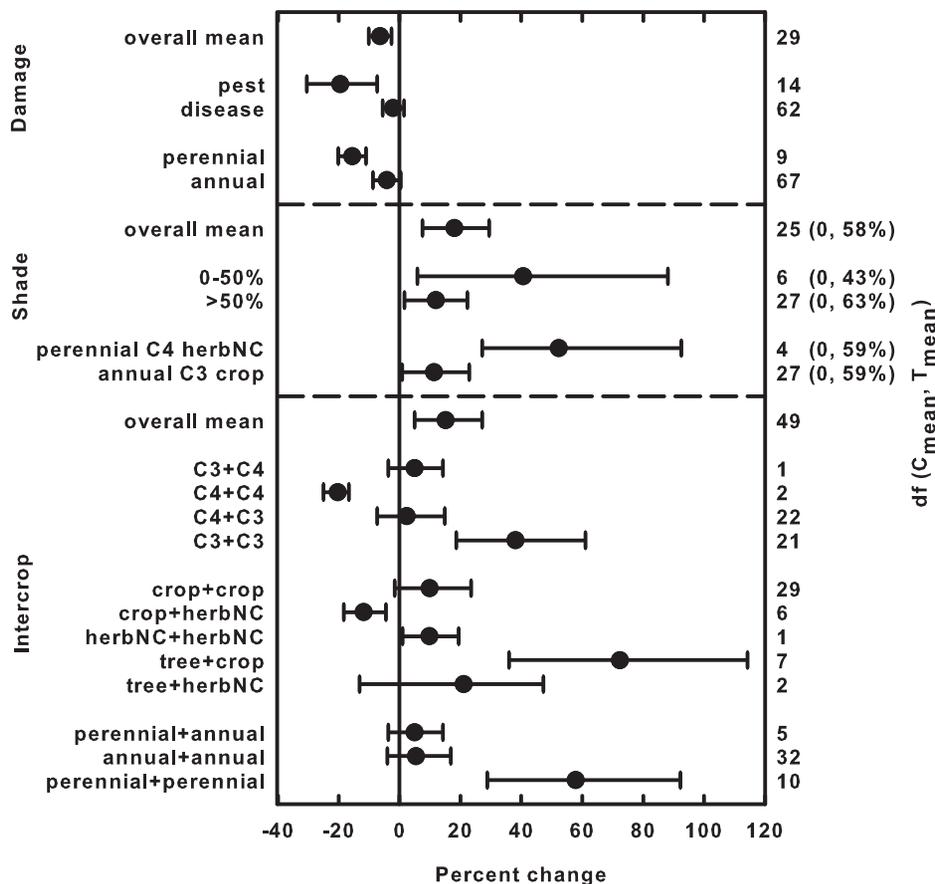


Fig. 4. Percentage change in ϵ_c due to treatments and categories within treatments in response to damage, shade, and intercropping. Symbols represent the mean percentage change from control conditions with 95% confidence intervals. Degrees of freedom (df) are listed on the right with mean control (C_{mean}) and treatment (T_{mean}) dosage in parentheses when applicable. Treatments are separated by dashed lines. herbNC = herbaceous non-crop species.

confounded. C_4 plants in which ϵ_c was enhanced by 52.3% were also perennial herbaceous non-crops and all C_3 plants in which ϵ_c was 11.2% greater were annual crops (Fig. 4).

Intercropping a commonly grown species with another species caused a significant stimulation (15.1%) in ϵ_c compared to growing that species alone (Fig. 4). The greatest stimulation within categorical variables came from the following categories: $C_3 + C_3$, tree + crop, and perennial + perennial (Fig. 4). However, there were a fair amount of data points that overlapped among those three categorical levels. The majority of tree + crop and perennial + perennial points were also $C_3 + C_3$ intercropping systems. Intercropping two C_4 plants actually decreased the ϵ_c response ratio (Fig. 4). This resulted in a 58% difference between C_4 and C_3 stands (Fig. 4). Intercropping usually indicates two crops being grown in the same space at the same time (Anil *et al.*, 1998), and unless the crops are the same size, one of them is likely to be shaded. Therefore, shade tolerance is preferred in the understory canopy. Unfortunately, the confounding factors in the shade treatment prevented any comparisons between categories that benefited from both shade and intercropping, but based on the shading effects, further experimentation could determine beneficial relationships in mixed stands containing plants of varying heights and shade tolerances to maximize ϵ_c on a land area basis. Another benefit of intercropping may come from nutrient sharing between legumes and non-legumes (Anil *et al.*, 1998). This was not supported by the analysis since none of the nitrogen fixation categories were significantly different (Supplementary Table S2), but it may be due to benefits being less evident when nutrients are not limiting (Anil *et al.*, 1998). Further tests with mixes of legumes and non-legumes on nutrient poor soils would be useful to determine the beneficial extent of legumes and their arrangement. The increases in ϵ_c may be due to additional intercropping advantages demonstrated by complementary crops, such as larger soil volume exploitation to gain

access to greater quantities of water and nutrients, improved pest/disease resistance, which directly influences ϵ_c (Fig. 4), and increased ground cover to improve soil conservation (Anil *et al.*, 1998).

Growth condition analyses revealed bias in ϵ_c measurements when plants were grown with limited rooting volume

Growth condition analyses yielded significant results within several treatments. Potted and greenhouse studies limited soil volume and exaggerated treatment effects on ϵ_c by 3-fold as compared to FACE and OTC experiments that studied plants grown directly in the field. While ϵ_c responded similarly to elevated $[CO_2]$ in FACE and OTC conditions, the ϵ_c of potted plants in OTCs demonstrated a 2-fold greater stimulation than in field OTC studies (Fig. 5). The depression of ϵ_c was 2-times greater in potted field experiments and 3-times greater in greenhouse studies versus water stress experiments conducted directly in the soil (Fig. 5). A greater than 5-fold increase in the response of ϵ_c to shade was demonstrated by potted studies compared to field-grown plant experiments (Fig. 5). Temperature treatments in the field significantly decreased ϵ_c compared to greenhouse studies (Fig. 5), but it should be noted that all sheltered field studies consisted of plants sown directly in the soil and treated with above optimal temperatures. All of these effects, except the temperature, suggest that soil volume affects ϵ_c responses to other treatments. Even under sufficient water conditions, plants in pots may experience decreased g_s and loss of leaf water potential due to imbalanced root to shoot ratios caused by root restriction (Tschaplinski and Blake, 1985). In elevated $[CO_2]$, plant canopies demonstrate an increased water use efficiency caused by decreased g_s (Leakey *et al.*, 2009), which may magnify the response of ϵ_c to elevated $[CO_2]$. The same concept may cause the greater decrease in ϵ_c

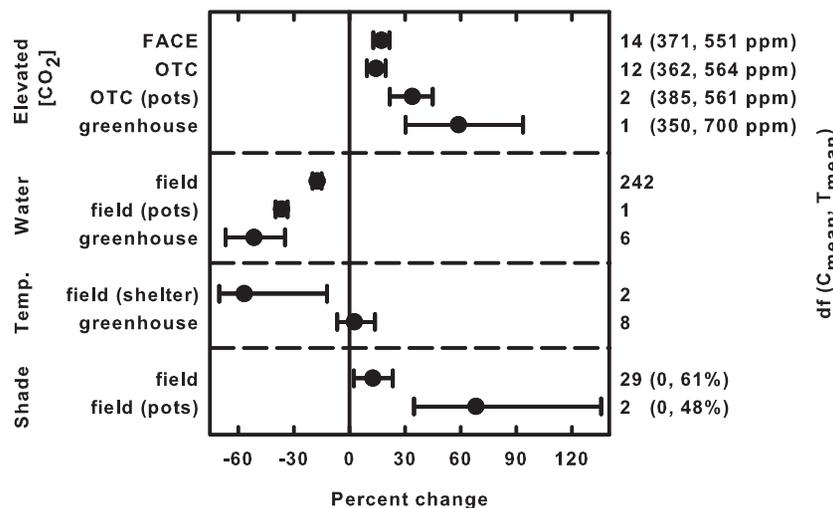


Fig. 5. Percentage change in ϵ_c due to growth condition within treatments in response to elevated $[CO_2]$, water, temperature, and shade. Symbols represent the mean percentage change from control conditions with 95% confidence intervals. Degrees of freedom (df) are listed on the right with mean control (C_{mean}) and treatment (T_{mean}) dosage in parentheses when applicable. Treatments are separated by dashed lines. FACE = free air concentration enrichment; OTC = open-top chamber; Temp. = temperature treatment.

with drought where decreased root surface area and drought conditions combine together to decrease ϵ_c . For plants already water limited, shading would provide a lesser need for cooling via transpiration and water loss, thereby conserving water longer. In the greenhouse, these effects may be enhanced by an altered environment such as lower incident light intensity and changes in relative humidity. With the exception of the temperature treatment, small sample sizes of non-field conditions compared to those of field conditions imply that these did not affect overall mean treatment effects. However, with the large-fold differences due to rooting volume conditions, trying to discern treatment effects from plants grown with limited soil volume would be misleading when attempting to determine the effects climate/weather and management cause in the field.

Conclusions

Using a novel approach to a meta-analytical study, this study statistically quantified how and to what extent ϵ_c is sensitive to several climate/weather factors and management practices that affect the ability of crops to close the yield gap between average and potential yields. Elevated $[\text{CO}_2]$ increases ϵ_c and therefore raises the ceiling on yield potential, but using biotechnology to engineer for greater responsiveness to elevated $[\text{CO}_2]$ will result in more substantial increases. Although shade also increases ϵ_c , the fact that it does so by decreasing S_i forecasts that it will not contribute as directly as elevated $[\text{CO}_2]$ to increasing yield potentials unless excess light is used more efficiently through intercropping, modified canopy architecture, or reduced chlorophyll content. The greatest ϵ_c response was seen with large quantities of added nitrogen, which also means severe decreases in ϵ_c could occur when nitrogen is limiting. Therefore, engineering plants for greater response to nitrogen applications is crucial to prevent decreases in ϵ_c and average yields. This is especially important in nutrient-poor areas predicted to suffer from worsening drought conditions since substantial decreases in legume productivity due to drought will negatively impact soil nitrogen availability for subsequent crops. Elevated $[\text{O}_3]$, water stress, temperature stress, and foliar damage depress measured yields and prevent gap closure, and these effects warrant advanced engineering for stress tolerance and agronomical practices such as intercropping to decrease pest and disease damage. Further experimentation to determine optimal practices is warranted, but growth condition analyses emphasize the importance of obtaining estimates of ϵ_c in field conditions for reliable results. Overall, these results indicate the significant extent to which environmental factors vary ϵ_c and support the concept that ϵ_c can be improved to increase yield potential and raise the ceiling to allow for greater rates of increase while closing the yield gap.

Supplementary material

Supplementary data are available at *JXB* online.

Supplementary Table S1. List of references containing data used in the meta-analysis.

Supplementary Table S2. Summary of treatment and categorical effects on the response variable (ϵ_c).

Supplementary Fig. S1. Regression analysis of percentage change in ϵ_c on nitrogen addition (kg ha^{-1}).

Acknowledgements

This work was supported by the United States Department of Agriculture/Agricultural Research Service. The authors acknowledge Courtney Leisner for her assistance during the process and all authors who contributed information upon request.

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