Photosynth Res (2016) 129:159–170
DOI 10.1007/s11120-016-0278-2

ORIGINAL ARTICLE

The effects of CO_2 and nutrient fertilisation on the growth and temperature response of the mangrove *Avicennia germinans*

Ruth Reef[1,2,3] · Martijn Slot[4] · Uzi Motro[5] · Michal Motro[6] · Yoav Motro[7] · Maria F. Adame[8] · Milton Garcia[4] · Jorge Aranda[4] · Catherine E. Lovelock[2] · Klaus Winter[4]

Received: 21 December 2015 / Accepted: 20 May 2016 / Published online: 3 June 2016

Abstract In order to understand plant responses to both the widespread phenomenon of increased nutrient inputs to coastal zones and the concurrent rise in atmospheric CO_2 concentrations, CO_2–nutrient interactions need to be considered. In addition to its potential stimulating effect on photosynthesis and growth, elevated CO_2 affects the temperature response of photosynthesis. The scarcity of experiments testing how elevated CO_2 affects the temperature response of tropical trees hinders our ability to model future primary productivity. In a glasshouse study, we examined the effects of elevated CO_2 (800 ppm) and nutrient availability on seedlings of the widespread mangrove *Avicennia germinans*. We assessed photosynthetic performance, the temperature response of photosynthesis, seedling growth and biomass allocation. We found large synergistic gains in both growth (42 %) and photosynthesis (115 %) when seedlings grown under elevated CO_2 were supplied with elevated nutrient concentrations relative to their ambient growing conditions. Growth was significantly enhanced under elevated CO_2 only under high-nutrient conditions, mainly in above-ground tissues. Under low-nutrient conditions and elevated CO_2, root volume was more than double that of seedlings grown under ambient CO2 levels. Elevated CO_2 significantly increased the temperature optimum for photosynthesis by ca. 4 °C. Rising CO_2 concentrations are likely to have a significant positive effect on the growth rate of *A. germinans* over the next century, especially in areas where nutrient availability is high.

Keywords Climate change · CO_2 · Eutrophication · Mangrove · Nitrogen · Phosphorus · Photosynthesis · RUBISCO · Temperature response · Tropics

✉ Ruth Reef
rr491@cam.ac.uk

1 Cambridge Coastal Research Unit, The University of Cambridge, Cambridge CB2 3EN, UK

2 School of Biological Sciences, The University of Queensland, St Lucia, QLD 4072, Australia

3 School of Earth, Atmosphere and Environment, Monash University, Clayton, VIC 3800, Australia

4 Smithsonian Tropical Research Institute, P.O. Box 0843-03092, Balboa, Ancon, Republic of Panama

5 Department of Ecology, Evolution and Behavior, Department of Statistics, The Federmann Centre for the Study of Rationality, The Hebrew University of Jerusalem, 91904 Jerusalem, Israel

6 The David Yellin Academic College of Education, 96342 Jerusalem, Israel

7 Plant Protection and Inspection Services, Ministry of Agriculture and Rural Development, 50250 Beit Dagan, Israel

8 Australian Rivers Institute, Griffith University, Nathan, QLD 4111, Australia

Introduction

Current increases in the concentration of CO_2 in the Earth's atmosphere are thought to have an overall positive effect on plant growth and productivity (Drake et al. 1997). However, due to factors interacting with CO_2, such as nutrient and water availability and temperature, measured growth responses to elevated CO_2 have often been variable (van der Sleen et al. 2015; Körner 2006; Ainsworth et al. 2003). In particular, progressive nitrogen limitation tends to reduce the long-term growth stimulation by elevated CO_2 (Luo et al. 2004; Reich et al. 2006; Norby et al. 2010), and thus under nutrient-limiting conditions, the stimulating

effects of elevated CO_2 on plant growth are often significantly reduced relative to nutrient-replete conditions (Oren et al. 2001). The handful of experiments studying the effects of elevated CO_2 (700–800 ppm) on mangrove seedlings have shown responses in growth and productivity, with a growth enhancement from 12 to up to 47 % under elevated CO_2 conditions (Reef et al. 2015; Farnsworth et al. 1996; McKee and Rooth 2008; Ball et al. 1997). Mangroves develop along tropical coastlines, where nutrients frequently are in low supply. In many mangrove forests, nitrogen and sometimes phosphorus have been shown to limit growth (Reef et al. 2010b) and saline conditions may be expected to limit responses to elevated CO_2 (Ball et al. 1997). Thus, to better understand the response of mangroves to elevated CO_2 conditions, CO_2–nutrient interactions need to be considered.

In addition to its potential stimulating effect on photosynthesis and growth, elevated CO_2 affects the temperature response of photosynthesis in C3 plants (Slot and Winter 2016). Since current mangrove distributions are strongly influenced by temperature (Duke et al. 1998; Hutchison et al. 2014; Woodroffe and Grindrod 1991; Quisthoudt et al. 2013), quantifying the effects of elevated CO_2 on the temperature response of mangroves is key to determining the fate of mangroves in the face of atmospheric and climate change. Photosynthesis is one of the most temperature-sensitive processes in plants (Berry and Björkman 1980). The carbon-fixing enzyme RUBISCO catalyses both carboxylation (and subsequently photosynthesis) and oxygenation (photorespiration) with CO_2 and O_2 as competing substrates. As temperatures rise, CO_2 solubility and the specificity of RUBISCO for CO_2 decrease to a greater extent than those of O_2 (Long 1991). Hence, the ratio between photorespiration and photosynthesis increases with increasing temperature (Jordan and Ogren 1984; Bernacchi et al. 2001), significantly reducing carbon assimilation rates and requiring higher CO_2 concentrations to attain similar levels of carbon assimilation. Based on theoretical models of photosynthesis, elevated CO_2 concentrations could have a strong effect on the temperature response of photosynthesis (Farquhar et al. 1980; Lloyd and Farquhar 2008), but experimental evidence for this is not well documented for tropical trees. A number of recent models predict a significant shift in mangrove distributions, for example the loss of mangrove forests from regions of high temperature and a reduction in productivity based on an anticipated rise in global temperature (Beaumont et al. 2011; Osland et al. 2013; Koch et al. 2015), but these predictions are based on the climatic niche of the present-day mangroves growing under current CO_2 concentrations. The scarcity of experiments testing how elevated CO_2 affects the temperature relationships of tropical trees hinders our ability to model how elevated CO_2 will affect primary productivity in these systems into the future (Cernusak et al. 2013).

Mangrove forests contribute a large proportion of the primary productivity on tropical coasts, which is important for carbon sequestration and support of both marine and terrestrial food webs (Duarte et al. 2013). Members of the genus *Avicennia* are dominant within higher latitude forests and are documented to have expanded their range in recent decades on three continents (Saintilan et al. 2014). Additionally, in the core of the mangrove distribution (tropical latitudes) they have an important role as they colonise sediments and are tolerant of disturbance (Fromard et al. 2004). In this study, we examined the effects of elevated CO_2 and nutrient availability on the mangrove *Avicennia germinans* (L.) L. We assessed the photosynthetic performance, the temperature response of photosynthesis, seedling growth and biomass allocation.

Methods

Avicennia germinans propagules were collected in July 2014 at Galeta Point, Panama (9°24′N, 79°51′W), and transferred to the Santa Cruz Experimental Field Facility, Smithsonian Tropical Research Institute, Gamboa, Panama (9°07′N, 79°42′W), where they were planted in individual 1.6-L tree pots (Short One Treepot™, 10 × 10 × 23 cm. Stuewe and Sons, Tangent, Oregon) filled with a mixture (50 %/50 %) of local clay-textured topsoil from a lowland previously forested private property and sand. The plants (propagules) were randomly assigned to one of two well-ventilated, naturally illuminated glasshouses receiving full sunlight ($n = 34$ pots per glasshouse), one with similar to ambient (ca. 400 ppm) CO_2 concentrations and one with an elevated (800 ppm) CO_2 concentration.

Elevated CO_2 was maintained by releasing CO_2 gas from a high-pressure cylinder in brief pulses to maintain CO_2 concentrations between 790 and 810 ppm. The glasshouses were equipped with split air conditioning units programmed to turn on when ambient air temperature exceeded 30 °C. Air temperature and relative humidity were recorded in the two glasshouses every 15 min using a data logger (CR10X; Campbell Scientific, Logan, Utah, USA). The conditions in each of the two glasshouses during the experiment are summarised in Table 1.

Seedlings were watered twice weekly with 300 ml salt solution that saturated the pots. Two nutrient treatments were implemented in each glasshouse, a low-nutrient treatment ($n = 17$ in each glasshouse) and a high-nutrient treatment ($n = 17$ in each glasshouse). The solution low in nutrients contained 0.06 mM KNO_3, 0.04 mM $Ca(NO_3)_2$, 0.01 mM $NH_4H_2PO_4$, 0.01 mM $(NH_4)_2HPO_4$, 0.01 mM $MgSO_4$, 2.5 μM H_3BO_3, 0.2 μM $MnSO_4$, 0.2 μM $ZnSO_4$,

Table 1 CO_2, temperature and humidity conditions in the two glasshouses between the 22nd of June and the 13th of October 2014

Parameters measured	Ambient CO_2 glasshouse	Elevated CO_2 glasshouse
Mean air temperature (°C) ± SD	28.6 ± 8.9	28.2 ± 3.4
Mean relative humidity (%) ± SD	67 ± 20	68 ± 22
Mean [CO_2] (ppm) ± SD	423 ± 17	827 ± 27

Measurements were taken every 5 min throughout the day

0.05 µM $CuSO_4$, 0.05 µM H_2MoO_4 and 2 µM $C_{10}H_{12}FeN_2NaO_8$ (ethylenediaminetetraacetic acid iron (III)-sodium salt), which is similar to the nutrient concentrations in mangrove porewater where they are not exposed to anthropogenic eutrophication (Alongi et al. 1993; Chen and Twilley 1999). The concentrations in the high-nutrient solution were five times those of the low-nutrient solution. Ocean salt (Instant Ocean, Blacksburg, VA, USA) was added to both nutrient solutions to a concentration of 20 g L^{-1}. Once a week, the plants received a rinse of fresh water (10 ml) from a spray bottle to simulate a rain event washing the salt from their leaves.

Two plants died during the experimental period. After 3 months of growth (October 6, 2014), photosynthetic temperature response curves were assessed for four randomly selected plants from each of the four treatments over the period of a week. All plants were harvested on the 14th of October 2014.

Photosynthetic temperature response curves

Photosynthetic gas exchange was measured on intact leaves of known area enclosed in a Walz gas-exchange cuvette with Peltier temperature control (GWK 3M Walz, Effeltrich, Germany) connected to an LI-6252 infrared gas analyser (Li-Cor, Lincoln NE, USA) under constant illumination of 1000 µmol m^{-2} s^{-1} from a red/blue LED light array. The CO_2 concentration of the air entering the chamber was set to 400 ppm for the seedlings grown at ambient CO_2 concentrations and to 800 ppm for the seedlings grown at elevated CO_2 concentrations. Following the enclosure of the leaf into the chamber, the chamber temperature was reduced to 20 °C for ~60 min. The temperature was then increased in 5 °C increments (every 20–30 min, when a stable reading was established) up to 40–50 °C. The youngest fully expanded leaves were studied. Four seedlings did not recover photosynthetic function following the enclosure into the chamber and removed from the analysis. Leaf temperature was measured using a copper–constantan thermocouple attached to the bottom surface of the leaf.

Temperature response data were fitted to the equation from Battaglia et al. (1996; Eq. 1) using the *nlsfit* function in R (Team 2014). The equation describes the photosynthetic rate (A) at a given temperature (T) as a parabolic relationship, with A_{opt} and T_{opt} being the maximal photosynthetic rate and the temperature at which A_{opt} is achieved, respectively.

$$A(T) = A_{opt} - b\left(T - T_{opt}\right)^2 \quad (1)$$

Analysis of variance was used to detect differences in the parameters A_{opt} (measured here as photosynthetic capacity, A_{max}), T_{opt} and the high-temperature CO_2 compensation point (where net CO_2 exchange is zero; T_{max}) among treatments.

Transpiration rate was calculated from the water vapour difference between the air leaving the chamber and the incoming air. Stomatal conductance at each temperature was calculated from the rate of transpiration divided by the leaf–air vapour pressure difference (D) in the air leaving the chamber relative to the incoming air. Intrinsic water use efficiency (WUEi) was calculated as the carbon assimilation rate divided by the stomatal conductance.

Plant growth parameters and elemental composition

Plant growth (stem length, no. of nodes, no. of leaves, no. of branches along the main stem) was monitored throughout the experiment. Leaf temperatures were measured for three leaves per seedlings 1 week prior to harvest on two cloudless days using an infrared thermometer. The measurements were repeated on all seedlings five times during the day (08:00, 10:00, 13:00, 16:00 and 20:00). Following the harvest, plants were divided into leaves, stem and roots. Leaves were kept in a sealed bag with a moist paper towel in order to maintain hydration status. Leaf area was measured using an LI-3100C leaf area meter (Li-Cor, Lincoln, NE, USA) and weighed. Washed roots free of soil were photographed against a dark background and analysed using the IJ Rhizo root analysis package (Pierret et al. 2013). The entire root system was measured for each seedling. Plant material was then washed in distilled water to remove external salt, patted dry and weighed after which it was dried at 70 °C for 5 days and reweighed.

Samples for leaf nutrient concentrations and isotopic composition were taken from finely ground leaves and roots from ten randomly selected plants from each treatment. All leaves from each plant were pooled before

grinding. The isotopic composition of the added CO_2 in the 800 ppm treatment differed slightly from that in ambient air. The correction for this was previously determined for this system by growing two C4 plants (*Saccharum spontaneum* and *Portulaca oleracea*) in the chambers. C4 plant variation in carbon discrimination in response to environmental factors is far smaller than that observed for C3 plants and can thus reflect the variation in the CO_2 isotopic composition of the air. A correction factor of 2 ‰ was used in foliar $\delta^{13}C$ values of seedlings from the 800 ppm treatment (Cernusak et al. 2011a).

Phosphorus (P) concentrations were determined using a colourimetric assay as described in Reef et al. (2010a). Leaf isotope values for $\delta^{13}C$ and $\delta^{15}N$ were measured from pooled samples of green leaves for ten seedlings from each treatment. Samples were measured in an elemental analyser isotope ratio mass spectrometer (EA-IRMS, Sercon System, Griffith University; analytical errors of 0.1 ‰ for $\delta^{13}C$ and 0.2 ‰ for $\delta^{15}N$). Nitrogen is expressed relative to atmospheric nitrogen and carbon relative to Vienna Pee-Dee Belemnite.

We used ANOVA to test for differences in growth parameters among the treatments. Root/shoot ratios were *logit* transformed prior to analysis. Partial correlation analysis was used to test the relationship between specific leaf area (SLA) and growth. Climate data for Galeta Point was downloaded from the Smithsonian Physical Monitoring Program climate station at the Galeta Marine Laboratory.

Results

Effects of CO_2 and soil nutrients on foliar physiology

Using a two-way ANOVA, we found significant effects of both CO_2 concentration and nutrient treatment on photosynthetic capacity, A_{max} (ANOVA, $F_{(1,11)} = 8.5$, $p = 0.014$, and $F_{(1,11)} = 5.6$, $p = 0.04$, respectively; Fig. 1a–d; Table 2), where A_{max} increases with increased CO_2 concentration and with nutrient enrichment, but more so when both elevated CO_2 and elevated nutrients were provided (Table 2).

Elevated CO_2 significantly increased the temperature optimum for photosynthesis by ca. 4 °C (ANOVA, $F_{(1,12)} = 17.3$, $p = 0.001$; Fig. 1a–d; Table 2). Despite the shift in the temperature optimum, the high-temperature CO_2 compensation point (T_{max}), i.e. the temperature at which net CO_2 exchange is zero, did not change significantly and was on average 41.8 (±3 SD) °C. The range of temperatures at which photosynthesis was near maximum (≥80 % of A_{max}) spanned 13 °C and shifted to higher temperatures with elevated CO_2 (Table 2).

Transpiration rate (E), stomatal water vapour conductance (Gs) and intrinsic water use efficiency (WUEi) are presented for leaf temperatures of 25 °C. Elevated CO_2 resulted in a significant reduction in stomatal conductance and transpiration relative to the ambient CO_2 treatment (ANOVA, $F_{(1,11)} = 5.7$, $p = 0.04$ and $F_{(1,10)} = 13.5$, $p = 0.004$; Fig. 2a, b, respectively), which contributed to a significant increase in WUEi ($F_{(1,10)} = 22.1$, $p < 0.001$; Fig. 2c), most notably under the high-nutrient regime ($p = 0.03$). The foliar $\delta^{13}C$ of leaves was significantly less negative in the elevated CO_2 treatment indicating that long-term water use efficiency (WUE) for the duration of the experiment was higher in this treatment ($F_{(1,35)} = 42.4$, $p < 0.001$; Fig. 2d).

There were no significant differences in leaf temperatures among the CO_2 and nutrient treatments. On sunny days, leaf temperatures of ambient CO_2-grown plants were found to be at the high range and sometimes exceeded the optimal temperature threshold for photosynthesis (defined here as the temperature range at which 80 % of maximum photosynthetic rates can be achieved, Table 2). For plants growing under elevated CO_2 conditions, leaf temperatures were well within the optimal range for photosynthesis (Table 2). Neither the CO_2 nor the nutrient treatment significantly affected leaf water content, which was on average (±SD) 71.3 % (±2.2 %) of the fresh weight.

Effects of CO_2 and nutrients on growth and biomass allocation

Seedling growth (total biomass accumulated) was significantly enhanced under elevated CO_2 but only under high-nutrient conditions (ANOVA $F_{(1,62)} = 9.2$, $p = 0.003$; Fig. 3a). In the high-nutrient treatment, the rise in CO_2 concentrations from 400 to 800 ppm resulted in a 44 % increase in biomass. Growth enhancement in the high-nutrient treatment occurred mainly in above-ground tissues (Fig. 3b), resulting in significantly lower root/shoot biomass ratios, with a more pronounced decrease in elevated CO_2-grown plants (ANOVA $F_{(1,62)} = 9.8$, $p = 0.003$). However, despite a lower overall allocation to roots versus shoots, root biomass under elevated CO_2 was significantly greater for the high relative to the low-nutrient treatment (ANOVA $F_{(1,62)} = 6.5$, $p = 0.013$; Fig. 3a). The increased allocation of biomass to shoots was associated with a significant increase in leaf area: for the high-nutrient treatment, elevated CO_2 resulted in a 55 % increase in leaf area and for the elevated CO_2 concentration, high-nutrient conditions resulted in a 71 % increase in leaf area (ANOVA $F_{(1,62)} = 13.9$, $p < 0.001$ and $F_{(1,62)} = 18.9$, $p < 0.001$, respectively; Fig. 3c),

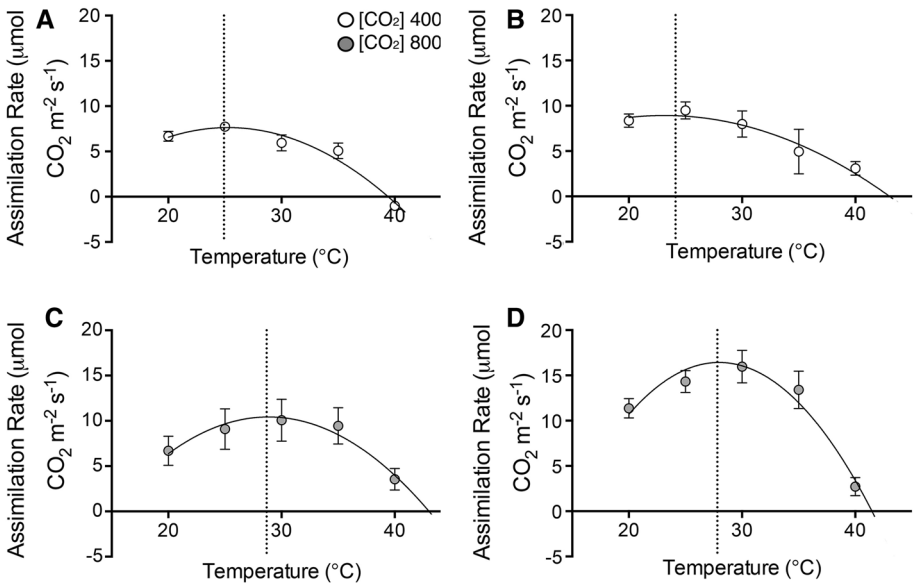

Fig. 1 Measured carbon assimilation rates **a** for attached, intact *Avicennia germinans* leaves as a function of leaf temperature in seedlings grown under (*top*) ambient (400 ppm) and (*bottom*) elevated (800 ppm) CO_2 concentrations subjected to low (*left*)- or high (*right*)-nutrient treatments. The measurements were made under saturating light conditions of 1000 µmol m^{-2} s^{-1}. *Points* are the mean (±SE) values for four seedlings, *fitted lines* are derived from the quadratic relationship described in Eq. 1, and *dotted vertical lines* denote the calculated temperature optimum for each treatment

Table 2 Mean (SD) values describing the temperature response of photosynthesis in *Avicennia germinans* seedling grown at ambient (ca. 400 ppm) and elevated (ca. 800 ppm) CO_2 concentrations and under two nutrient regimes (low and high)

Parameter	CO_2 ppm Nutrients			
	400 Low	400 High	800 Low	800 High
A_{max} (µmol cm^{-2} s^{-1})	7.5 (1.5)[a]	9.4 (1.4)[b]	10.3 (4.4)[c]	16.1 (3.6)[d]
T_{opt} (°C)	24.9 (1.6)[a]	24.1 (2.9)[a]	28.7 (1.8)[b]	27.8 (0.6)[b]
T_{max} (°C)	39.4 (0.6)[a]	41.6 (5.5)[a]	43.8 (2.2)[a]	42.2 (1.9)[a]
$T_{80\ \%\ Amax}$ (°C)	19.0–31.9	17.2–30.9	22.4–35.3	21.8–34.4

A_{max} is the maximal carbon assimilation rate at light saturation and T_{opt} is the temperature at which A_{max} is achieved. T_{max} is the temperature at which the upper CO_2 compensation point occurs, above which net CO_2 loss occurs. Values were calculated from the quadratic relationship fit to the temperature series from each seedling (Eq. 1). $N = 4$ seedlings per treatment. Different letters indicate significant differences among the treatments ($p < 0.05$)

In contrast, in the low-nutrient treatment, elevated CO_2 did not lead to significant biomass gains (Tukey HSD, $p = 0.96$). Increasing nutrient concentrations fivefold alone did not lead to significant biomass gains at ambient CO_2 levels.

Using partial correlation (while controlling for nutrient treatment and CO_2 concentration), we found specific leaf area (SLA) to be correlated with relative growth rate, RGR ($R = -0.47$, $p < 0.001$; Fig. 3d), and thus higher growth rates were associated with lower SLA values. The slope of this relationship was independent of nutrient treatment or CO_2 concentration ($p > 0.05$).

Consistent with the stimulation of biomass growth, seedlings in the high nutrient–elevated CO_2 treatment had longer stems and more leaves than seedlings from other treatments (ANOVA, $F_{(1,62)} = 4.7$, $p = 0.03$ and $F_{(1,62)} = 7.0$, $p = 0.01$, respectively, Table 3). Notwithstanding the difference in size, we did not observe changes to growth allocation patterns in these stems (e.g. branching rates and internode lengths did not differ among treatments, Table 3).

Root structure was significantly influenced by the CO_2 and nutrient treatments. Roots were significantly longer in elevated CO_2-grown seedlings relative to those grown under ambient CO_2 ($F_{(1,37)} = 9.5, p = 0.004$). Under low-nutrient conditions and elevated CO_2, root volume was more than double that of seedlings grown under ambient CO_2 levels ($F_{(1,37)} = 5.8, p = 0.02$, Table 3). Mean root diameter was also affected, with a higher frequency of fine roots in the ambient CO_2–low nutrient and high CO_2–high nutrient treatments ($F_{(1,37)} = 28.4, p < 0.001$, Table 3). We identified three major root types in our seedlings: fine roots with diameters < 2 mm, lateral roots (diameter = 2–4 mm) and pneumatophores, which developed in a few seedlings (diameter > 4 mm). Fine roots made up on average 76 % of the total root length. The fine root ratio (fine roots/total root biomass) was higher in the low-nutrient treatment under ambient CO_2 conditions, as was the total fine root length.

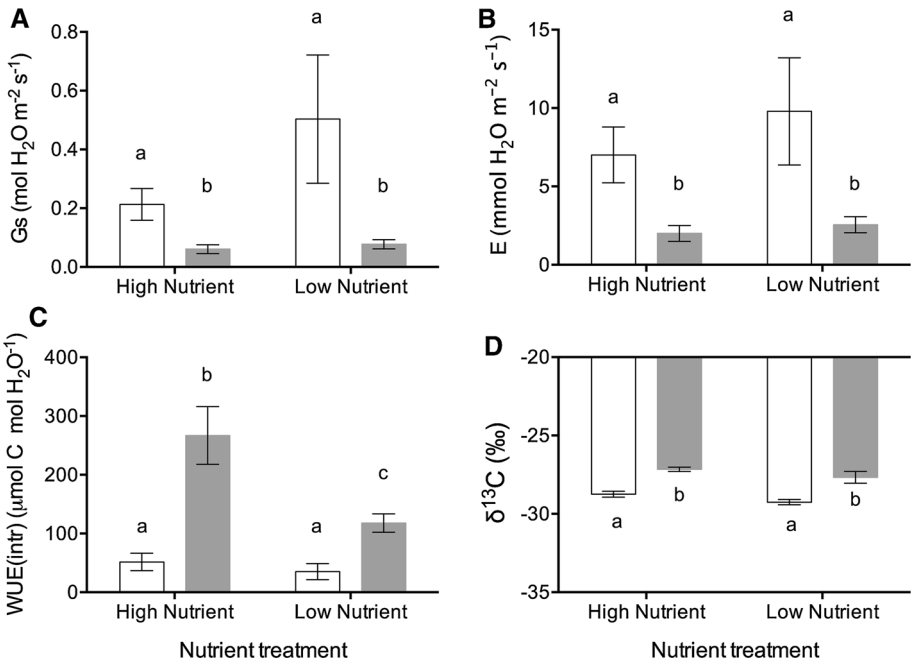

Fig. 2 Mean (±SE) **a** stomatal water vapour conductance (Gs), **b** transpiration (E) and **c** intrinsic water use efficiency (WUEi) of attached, intact leaves of four seedlings from each treatment at a leaf temperature of 25 °C, irradiance of 1000 µmol m^{-2} s^{-1} and CO_2 concentrations of 400 ppm for seedlings from the low- and high-nutrient treatments grown at ambient CO_2 levels (*open bars*), or 800 ppm for seedlings grown at the elevated CO_2 concentration (*filled bars*). **d** Foliar $\delta^{13}C$ values for $N = 10$ seedlings from each treatment measured at the end of the experiment. *Different letters* denote significant differences among treatments ($p < 0.05$)

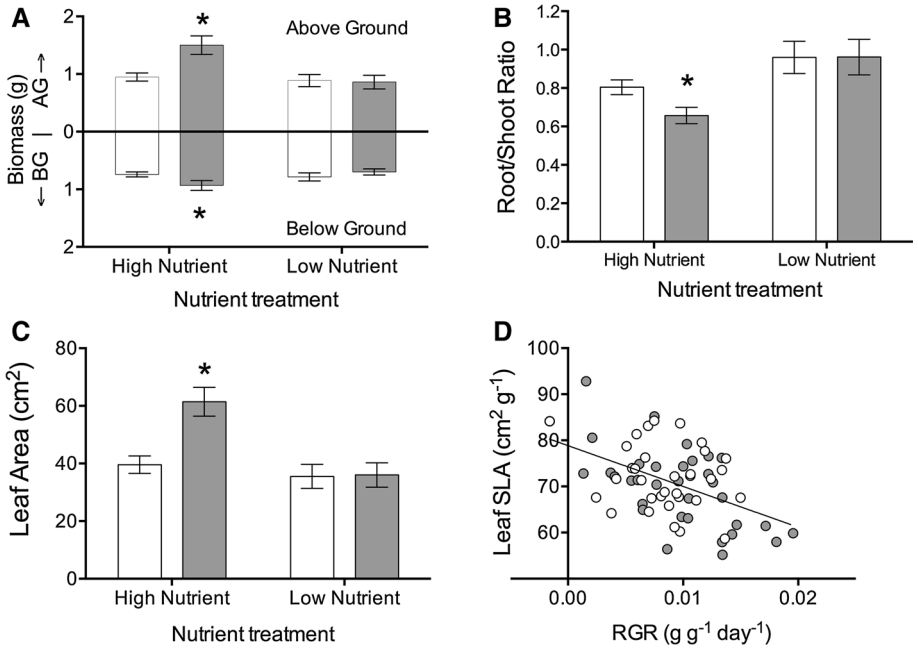

Fig. 3 Mean (±SE) **a** final above-ground (AG) and below-ground (BG) biomass, **b** root/shoot biomass ratio and **c** total leaf area of seedlings grown under ambient (400 ppm, *open bars*) or elevated (800 ppm, *filled bars*) CO_2 concentrations and subject to either a low- or high-nutrient treatment. $N = 16$–17 seedlings per treatment. *Asterisk* denotes significant differences among treatments ($p < 0.05$). **d** Relationship between relative growth rate (RGR) and mean specific leaf area (SLA) for each seedling. The fitted linear regression is of the form $SLA = -885RGR + 78.9$ ($R^2 = 0.22$, $p < 0.001$). *Open* and *filled circles* represent seedlings grown under ambient or elevated CO_2 concentrations, respectively

Under elevated CO_2 conditions, the effect of nutrients on fine root production was reversed, with a significant decrease in the fine root ratio in the low-nutrient treatment. However, total fine root length remained higher in elevated CO_2 than under ambient CO_2 conditions for both nutrient treatments. Roots from elevated CO_2-grown seedlings also had higher concentrations of carbon, regardless of the nutrient treatment ($F_{(1,36)} = 15.5$, $p < 0.001$, Table 4).

Table 3 Mean (SD) values describing the morphological response of *Avicennia germinans* seedlings to ambient (ca. 400 ppm) and elevated (ca. 800 ppm) CO_2 concentrations and two nutrient regimes (low and high)

Parameter	CO_2 ppm Nutrients			
	400 Low	400 High	800 Low	800 High
Stem length (cm)	17.4 (5.8)^a	17.6 (4.6)^a	14.6 (4.8)^a	20.2 (5.0)^b
Internode length (cm)	3.2 (0.8)^a	3.1 (0.8)^a	2.9 (0.7)^a	3.0 (0.7)^a
Leaves per seedling	9.8 (4.5)^a	11.2 (5.8)^a	8.5 (3.7)^a	12.9 (3.5)^b
Branching rate (cm^{-1})	0.10 (0.07)^a	0.10 (0.09)^a	0.09 (0.05)^a	0.08 (0.04)^a
Leaf mortality rate (day^{-1})	0.03 (0.03)^a	0.01 (0.02)^b	0.03 (0.03)^a	0.01 (0.02)^b
Root length (cm)	864.8 (307.7)^a	654.6 (249.2)^a	1065.2 (446.9)^b	1242.9 (585.7)^b
Root volume (cm^3)	2.1 (1.5)^a	3.7 (1.9)^a	4.5 (2.0)^b	3.4 (1.7)^a
Mean root diameter (mm)	0.56 (0.13)^a	0.90 (0.12)^b	0.80 (0.06)^b	0.66 (0.17)^a
Fine root length ratio	0.85 (0.08)^a	0.68 (0.08)^b	0.73 (0.05)^b	0.80 (0.1)^a

$N = 17$ seedlings per treatment for above-ground measurements and $N = 10$ per treatment for root analysis. Different letters indicate significant differences among the treatments ($p < 0.05$)

Table 4 Mean (SD) values describing the elemental composition of roots and leaves of *Avicennia germinans* seedlings grown at ambient (ca. 400 ppm) and elevated (ca. 800 ppm) CO_2 concentrations and two nutrient regimes (low and high)

Parameter	CO_2 ppm Nutrients			
	400 Low	400 High	800 Low	800 High
Leaves				
%C	39.5 (0.9)^a	39.5 (0.5)^a	39.5 (0.7)^a	39.7 (0.6)^a
%N	3.8 (0.7)^a	3.5 (0.6)^a	3.7 (0.7)^a	3.7 (0.4)^a
%P	0.51 (0.60)^a	0.51 (0.70)^a	0.44 (0.8)^b	0.47 (0.11)^b
C:N	10.7 (1.8)^a	11.5 (1.9)^a	11.1 (2.4)^a	10.9 (1.2)^a
C:P	78.3 (12.6)^a	78.3 (11.3)^a	92.1 (17.1)^b	88.7 (19.6)^b
N:P	7.45 (1.6)^a	6.86 (2.0)^a	8.41 (2.3)^a	7.87 (2.1)^a
N_a ($g\ m^{-2}$)	0.53 (0.1)^a	0.47 (0.1)^a	0.54 (0.2)^a	0.53 (0.1)^a
P_a ($g\ m^{-2}$)	0.07 (0.01)^a	0.07 (0.01)^a	0.06 (0.01)^a	0.07 (0.01)^a
Roots				
%C	31.1 (2.2)^a	31.3 (2.1)^a	33.7 (2.1)^b	33.9 (1.8)^b
%N	1.0 (0.04)^a	1.2 (0.1)^b	1.1 (0.1)^a	1.3 (0.1)^c
%P	0.50 (0.05)^a	0.50 (0.08)^a	0.62 (0.14)^a	0.58 (0.1)^a
C:N	30.3 (2.6)^a	26.7 (2.1)^b	30.5 (2.7)^a	26.7 (2.7)^b
C:P	63.1 (5.2)^a	65.0 (13.0)^a	56.2 (11.2)^a	60.0 (9.6)^a
N:P	2.0 (2.0)^a	2.44 (0.7)^b	1.77 (0.38)^a	2.24 (4.1)^b

$N = 10$ seedlings per treatment for above-ground measurements and N = 10 per treatment for root analysis. Different letters indicate significant differences among the treatments ($p < 0.05$)

Effects of CO_2 and nutrients on plant nutrient content

Phosphorus (P) concentrations in plant tissues were significantly affected by the CO_2 treatment. Elevated CO_2 seedlings had significantly higher concentration of P in their root tissues, relative to ambient CO_2-grown seedlings (ANOVA $F_{(1,36)} = 11.5$, $p = 0.002$, Table 4). In leaves, we found the opposite, lower P concentrations in seedlings from the elevated CO_2 treatment relative to ambient CO_2 ($F_{(1,35)} = 5.1$, $p = 0.03$, Table 4). The nutrient treatment had no significant effect on tissue P concentrations.

The exhaustion of maternal nutrient reserves as the seedlings matured led to a significant loss of foliage in low nutrient-grown seedlings where leaf mortality rates were more than double those of the high nutrient-grown seedlings (ANOVA, $F_{(1,62)} = 4.8$, $p = 0.03$, Table 3). However, N or P concentrations in leaves of the low-nutrient plants were not significantly lower than those in plants from the high-nutrient treatment (Table 4). Differences in

elemental composition between the nutrient treatments were detected in the roots, with higher %N, lower C:N and higher N:P in the high-nutrient plants ($F_{(1,36)} = 24.8$, $p < 0.001$, $F_{(1,36)} = 7.2$, $p = 0.01$ and $F_{(1,36)} = 21$, $p < 0.001$, respectively, Table 4).

Discussion

We found large synergistic gains in both photosynthesis and growth in *A. germinans* seedlings when seedlings grown under elevated CO_2 were supplied with elevated nutrient concentrations. In the high nutrient–elevated CO_2 treatment, photosynthesis was enhanced on average by 75 % relative to the high nutrient–ambient CO_2 grown seedlings, and 115 % when compared with the low nutrient–ambient CO_2 grown seedlings. Growth was enhanced by 42 % in the elevated CO_2–high nutrient treatment relative to ambient CO_2–high nutrient seedlings. As has been observed in other species, growth was less sensitive than photosynthesis to elevated CO_2 (Kirschbaum 2011). Despite significant differences in WUEi and WUE among the nutrient and CO_2 treatments, plant WUEi was not associated with growth or productivity. This is consistent with growing evidence that indicates that mangrove growth is not limited by water availability at moderate salinities (Reef et al. 2012).

Elevated CO_2 had a significant effect on the temperature dependence of light-saturated photosynthesis as is predicted by theoretical models (Farquhar et al. 1980; Lloyd and Farquhar 2008). The optimal temperature for carbon fixation increased from 24.5 °C at the CO_2 concentrations of 400 ppm to 28.3 °C in plants that were grown and measured at 800 ppm CO_2, an increase of nearly 4 °C, which is higher than the predicted increase in mean global temperature for 2100 for moderate emissions scenarios (IPCC 2013). The effect of elevated CO_2 on the temperature response of photosynthesis has been shown to be robust over time for a number of species grown in the Nevada FACE facility (Taub et al. 2000). T_{max}, the temperature at which net assimilation is zero, was not significantly affected by elevated CO_2 concentrations, remaining on average 41.8 °C. Irreversible damage in tropical tree leaves has been shown to occur at temperatures >50 °C (Krause et al. 2010, 2014) so it is unlikely that T_{max} represents the point where damage to the photosynthetic apparatus occurs.

Despite differences in transpiration rates of 74 % among the different CO_2 and nutrient treatments, leaf temperatures measured during the experiment were not significantly higher in the elevated CO_2-grown seedlings. This could be due to the fact that transpiration plays a relatively small role in leaf temperature regulation compared to the important influence of air temperature and irradiance (Miller 1972). This may be especially true in mangroves, where non-evaporative cooling strategies (e.g. leaf orientation, pubescence and salt excretion) are adaptations that maintain high water use efficiencies in these species (reviewed in Reef and Lovelock 2014b). It should also be noted that air movement inside the glasshouse increased the coupling between air and leaf temperatures.

The photosynthesis temperature response measured for *A. germinans* was of similar shape to the temperature response measured for the congeneric *Avicennia marina* (Ball et al. 1988), and while T_{opt} of *A. germinans* was 3 °C lower than that of its Australian counterpart, the high-temperature CO_2 compensation point (the temperature at which net CO_2 exchange is zero, T_{max}) was similar to that of *A. marina*. Evidence from field measurements suggests that photosynthesis in *Bruguiera parviflora* from northern Queensland was strongly depressed at leaf temperatures >34 °C (Cheeseman et al. 1991). Also in northern Queensland, assimilation rates in *Rhizophora stylosa* decreased linearly as temperatures increased from 27 to 40 °C and was at nearly the CO_2 compensation point at 39.5 °C (Andrews and Muller 1985). However, in both these studies, the effect of temperature on carbon assimilation rates was confounded by coinciding changes in light levels, humidity and differences in leaf angles. The CO_2 compensation point (T_{max}) for *A. germinans* in our study was on average 41.8 ± 3 °C, and while we found a significant increase in T_{opt} with elevated CO_2, we did not find an increase in the high temperature threshold for this species under elevated CO_2 conditions.

The optimal temperature for photosynthesis under ambient CO_2 conditions was lower than the T_{leaf} measured for the seedlings throughout the day (Fig. 1). T_{opt} was also lower than the mean temperature in the glasshouse (Table 1) and lower than the mean daily atmospheric temperature recorded at Punta Galeta, where the plant material was collected, in the years 2002–2015 between 07:00 and 16:00 (27.8 °C ± 2). However, the temperature range of near-optimal photosynthetic performance of the seedlings was very broad (approx. 13 °C, Table 2) and the leaf temperatures measured in the glasshouse during growth were within this range. Woody evergreen plants have a higher temperature homeostasis of photosynthesis and alter T_{opt} in response to the growing temperature to a lesser extent than other vegetation groups (Yamori et al. 2013). Nonetheless, a T_{leaf} that is on average higher than T_{opt} suggests an incomplete acclimation to the mean growing temperature. It is possible that broad response of photosynthesis to temperature in *A. germinans* reflects its broad latitudinal distribution. A broad temperature tolerance was also found in the photosynthesis of the congeneric *A. marina* (Ball et al. 1988), which also has broad

latitudinal distribution. There is growing evidence that not all plant species are capable of complete photosynthetic thermal acclimation to growth temperature (e.g. Dillaway and Kruger 2010). Our findings for *A. germinans* support this possibility. Relatively low T_{opt} compared to mean daily temperature may also indicate acclimation of photosynthesis to early morning conditions when the majority of photosynthetic carbon gain in this species occurs (Smith et al. 1989). The mean temperature in the early morning (06:00–09:00) at Punta Galeta was (26.7 ± 1.9). In mangroves, midday depressions in photosynthesis are common (Björkman et al. 1988; Andrews and Muller 1985; Cheeseman et al. 1991), with some field studies showing a peak in photosynthesis before 08:00 AM and a cessation of photosynthesis by 11:00 AM (Cheeseman et al. 1991). An incomplete acclimation to high ambient temperatures could be one of the causes of these depressions. Photosynthesis temperature response in three Australian mangrove species (*Bruguiera gymnorrhiza, Rhizophora apiculata and Avicennia marina*), measured under ambient (unspecified) CO_2 concentrations, showed a broad temperature optimum (25–30 °C), which was significantly lower than the leaf temperatures measured on sun-exposed leaves as early as 08:25 AM (Ball et al. 1988). In the Ball et al.'s (1988) study, it was shown that leaf angle in mangroves is optimised to reduce leaf temperatures rather than maximise light capture, resulting in lower rates of photosynthesis. Irrespective of the underlying pressure that leads to selection for the broad temperature optima of photosynthesis and the cause of incomplete acclimation to the mean growing temperature, the increase in T_{opt} with increasing CO_2 concentrations could result in improved photosynthetic performance and growth rates for this species within the tropics as CO_2 concentrations continue to increase.

Low nutrient availability restricted the growth response of the mangrove *A. germinans* to elevated CO_2 despite significant improvements to photosynthesis and water use efficiency. The additional carbon fixed in the elevated CO_2–low nutrient treatment did not contribute to additional biomass and it is thus possible that night-time respiration rates in this treatment were higher than those in the ambient CO_2–low nutrient treatment. Higher rates of dark respiration in leaves of plants grown under elevated CO_2 conditions were reported for a variety of species, in part as a result of higher mitochondria densities (Griffin et al. 2001; Leakey et al. 2009). Elevated CO_2 stimulated growth mainly above ground (increasing leaf area), although significant increases in below-ground biomass were also detected relative to ambient CO_2 concentrations. Elevated CO_2 did not directly affect SLA, suggesting that excess carbon accumulation (which could lead to a downregulation of photosynthesis) did not occur in the leaves of plants from the elevated CO_2 treatment. The enhancements we observed in plant performance with elevated CO_2 are consistent with previous studies conducted in greenhouses with mangrove seedlings (Farnsworth et al. 1996; Reef et al. 2015; McKee and Rooth 2008; Ball et al. 1997) and other plant species (Winter et al. 2001a, b; Ainsworth and Long 2005).

The combination of elevated CO_2 and elevated nutrients resulted in significantly higher leaf areas but no significant differences in the nitrogen and carbon concentration of leaves. An analysis of 16 FACE experiments worldwide found no effects of elevated CO_2 on foliar nitrogen concentrations in woody plants (Nowak et al. 2004). However, due to the increase in leaf area, an increase in nitrogen uptake did occur at the whole-plant level. Elevated CO_2 led to a reduction in foliar phosphorus concentrations (Table 4), a phenomenon which has been observed previously in *A. germinans* (Reef et al. 2015). This could be due to reduced transpiration rates (Fig. 3b), possibly involving subsequent lower translocation rates of P to the shoot via the xylem stream, as has been suggested for other tropical trees (Cernusak et al. 2011b). This is further supported by the increase in P concentrations (and small increases in %N) in the roots of the elevated CO_2 seedlings (Table 4). The reduction in foliar phosphorus concentrations under elevated CO_2 was tended to be less pronounced in the high-nutrient treatment. Elevated CO_2-induced reduction in whole seedling transpiration rates could thus have a significant effect on growth rates in mangrove forests where P is the limiting nutrient for growth such as in forests that are hydrologically isolated from regular tidal inundation (Feller et al. 2003).

Elevated CO_2 had a significant effect on roots, increasing root length and biomass and also the carbon concentration in the roots, but did not increase allocation of biomass to roots (except under high nutrient levels) as has been shown in other woody species (Hättenschwiler and Körner 1997). Root morphology was influenced in a complex interaction between elevated CO_2 and nutrient availability as root systems under elevated CO_2 and high-nutrient conditions tended to have a lower proportion of biomass allocated to roots, but roots had a higher proportion of fine roots (Fig. 3; Table 3). The increase in fine root production we observed for *A. germinans* under elevated CO_2 conditions is consistent with allocation models based on findings from other tree species (Dybzinski et al. 2015) and is suggested to be driven by the use of carbon exudates to prime microbial populations to enhance N release for plant growth (Phillips et al. 2011). Root development is influenced by complex interactions among nutrient and water demands of the shoot (Poorter et al. 2012) and carbohydrate availability (Eveland and Jackson 2012). Reduction in transpiration in seedlings grown under elevated CO_2 (and increased WUE) reduces the demand for water, which may be balanced by an increase in nutrient

demand due to higher growth rates (Chapin 1980), leading to little overall change in allocation to roots under low-nutrient conditions (Fig. 3). As the rate of root development in mangroves is an important determinant of seedling establishment success in the soft sediment of tidal flats (Balke et al. 2011), the rapid elongation of roots under elevated CO_2 may increase survivorship of seedlings. Potential changes under elevated CO_2 in allocation to root biomass, or alterations to root morphology and elemental composition, which may influence decomposition, are important in mangrove forests as these factors are likely to influence capacity for carbon sequestration in these habitats and their responses to sea-level rise (Krauss et al. 2014).

Mangroves in a changing environment

Rising CO_2 concentrations are likely to have a significant positive effect on the growth rate of the widespread mangrove *A. germinans* over the next century, especially in areas where nutrient availability is high. For a congenitor in the Pacific Ocean (*A. marina*), there is evidence that primary production has already been influenced by elevated CO_2 (Reef and Lovelock 2014a). Increased nutrient loading in coastal areas is widespread and synergistic interactions with elevated CO_2 are likely to result in overall increases in mangrove biomass, C sequestration and belowground C storage. Elevated CO_2 concentrations will affect the temperature response of photosynthesis in this species more so than the predicted rise in mean global temperature over this period (2.5–3 °C), possibly mitigating growth inhibition by future high-temperature anomalies.

Acknowledgements We would like to thank Dr Aurelio Virgo for technical support. Funding for this study was provided by an Australian Research Council Discovery Early Career Research Award to RR (DE120101706) and a Marie Curie Fellowship to RR (FP7-623720—STORM). Propagules were collected under Autoridad Nacional del Ambiente, Panama scientific permit No. SC/P-7-14. All data used in this manuscript are presented in the manuscript.

References

Ainsworth EA, Long SP (2005) What have we learned from 15 years of free-air CO_2 enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy properties and plant production to rising CO_2. New Phytol 165(2):351–372. doi:10.1111/j.1469-8137.2004.01224.x

Ainsworth EA, Rogers A, Blum H, Nösberger J, Long SP (2003) Variation in acclimation of photosynthesis in *Trifolium repens* after eight years of exposure to Free Air CO_2 Enrichment (FACE). J Exp Bot 54(393):2769–2774. doi:10.1093/jxb/erg309

Alongi DM, Christoffersen P, Tirendi F (1993) The influence of forest type on microbial-nutrient relationships in tropical mangrove sediments. J Exp Mar Biol Ecol 171(2):201–223. doi:10.1016/0022-0981(93)90004-8

Andrews TJ, Muller GJ (1985) Photosynthetic gas exchange of the mangrove, *Rhizophora stylosa* Griff., in its natural environment. Oecologia 65(3):449–455. doi:10.1007/BF00378922

Balke T, Bouma T, Horstman E, Webb E, Erftemeijer P, Herman P (2011) Windows of opportunity: thresholds to mangrove seedling establishment on tidal flats. Mar Ecol Prog Ser 440:1–9

Ball MC, Cowan IR, Farquhar GD (1988) Maintenance of leaf temperature and the optimisation of carbon gain in relation to water loss in a tropical mangrove forest. Aust J Plant Physiol 15:263–276

Ball MC, Cochrane MJ, Rawson HM (1997) Growth and water use of the mangroves *Rhizophora apiculata* and *R. stylosa* in response to salinity and humidity under ambient and eievated concentrations of atmospheric CO_2. Plant Cell Environ 20:1158–1166

Battaglia M, Beadle C, Loghhead S (1996) Photosynthetic temperature responses of *Eucalyptus globulus* and *Eucalyptus nitens*. Tree Physiol 16:81–89

Beaumont LJ, Pitman A, Perkins S, Zimmermann NE, Yoccoz NG, Thuiller W (2011) Impacts of climate change on the world's most exceptional ecoregions. Proc Natl Acad Sci 108(6):2306–2311. doi:10.1073/pnas.1007217108

Bernacchi CJ, Singsaas EL, Pimentel C, Portis AR Jr, Long SP (2001) Improved temperature response functions for models of Rubisco-limited photosynthesis. Plant Cell Environ 24(2):253–259. doi:10.1111/j.1365-3040.2001.00668.x

Berry J, Björkman O (1980) Photosynthetic response and adaptation to temperature in higher plants. Annu Rev Plant Physiol 31(1):491–543. doi:10.1146/annurev.pp.31.060180.002423

Björkman O, Demmig B, Andrews T (1988) Mangrove photosynthesis: response to high-irradiance stress. Funct Plant Biol 15(2):43–61. doi:10.1071/PP9880043

Cernusak LA, Winter K, Martínez C, Correa E, Aranda J, Garcia M, Jaramillo C, Turner BL (2011a) Responses of legume versus nonlegume tropical tree seedlings to elevated CO_2 concentration. Plant Physiol 157(1):372–385. doi:10.1104/pp.111.182436

Cernusak LA, Winter K, Turner BL (2011b) Transpiration modulates phosphorus acquisition in tropical tree seedlings. Tree Physiol 31(8):878–885. doi:10.1093/treephys/tpr077

Cernusak LA, Winter K, Dalling JW, Holtum JAM, Jaramillo C, Körner C, Leakey ADB, Norby RJ, Poulter B, Turner BL, Wright SJ (2013) Tropical forest responses to increasing atmospheric CO_2: current knowledge and opportunities for future research. Funct Plant Biol 40:531–551

Chapin FS (1980) The mineral nutrition of wild plants. Annu Rev Ecol Syst 11:233–260

Cheeseman JM, Clough BF, Carter DR, Lovelock CE, Eong O, Sim RG (1991) The analysis of photosynthetic performance in leaves under field conditions: a case study using *Bruguiera* mangroves. Photosynth Res 29(1):11–22. doi:10.1007/BF00035202

Chen R, Twilley R (1999) Patterns of mangrove forest structure and soil nutrient dynamics along the Shark River estuary, Florida. Estuaries 22(4):955–970. doi:10.2307/1353075

Dillaway DN, Kruger EL (2010) Thermal acclimation of photosynthesis: a comparison of boreal and temperate tree species along a latitudinal transect. Plant Cell Environ 33(6):888–899. doi:10.1111/j.1365-3040.2010.02114.x

Drake BG, Gonzàlez-Meler MA, Long SP (1997) More efficient plants: a consequence of rising atmospheric CO_2? Annu Rev Plant Physiol Plant Mol Biol 48(1):609–639. doi:10.1146/annurev.arplant.48.1.609

Duarte CM, Losada IJ, Hendriks IE, Mazarrasa I, Marba N (2013) The role of coastal plant communities for climate change mitigation and adaptation. Nat Clim Change 3(11):961–968. doi:10.1038/nclimate1970

Duke NC, Ball MC, Ellison JC (1998) Factors influencing biodiversity and distributional gradients in mangroves. Glob Ecol Biogeogr Lett 7(1):27–47. doi:10.2307/2997695

Dybzinski R, Farrior CE, Pacala SW (2015) Increased forest carbon storage with increased atmospheric CO_2 despite nitrogen limitation: a game-theoretic allocation model for trees in competition for nitrogen and light. Glob Change Biol 21(3):1182–1196. doi:10.1111/gcb.12783

Eveland AL, Jackson DP (2012) Sugars, signalling, and plant development. J Exp Bot 63(9):3367–3377. doi:10.1093/jxb/err379

Farnsworth EJ, Ellison AM, Gong WK (1996) Elevated CO_2 alters anatomy, physiology, growth, and reproduction of red mangrove (*Rhizophora mangle* L.). Oecologia 108(4):599–609. doi:10.1007/bf00329032

Farquhar GD, von Caemmerer S, Berry JA (1980) A biochemical model of photosynthetic CO_2 assimilation in leaves of C3 species. Planta 149(1):78–90. doi:10.1007/BF00386231

Feller IC, McKee KL, Whigham DF, O'Neill JP (2003) Nitrogen vs. phosphorus limitation across an ecotonal gradient in a mangrove forest. Biogeochemistry 62:145–175

Fromard F, Vega C, Proisy C (2004) Half a century of dynamic coastal change affecting mangrove shorelines of French Guiana. A case study based on remote sensing data analyses and field surveys. Mar Geol 208(2–4):265–280. doi:10.1016/j.margeo.2004.04.018

Griffin KL, Anderson OR, Gastrich MD, Lewis JD, Lin G, Schuster W, Seemann JR, Tissue DT, Turnbull MH, Whitehead D (2001) Plant growth in elevated CO_2 alters mitochondrial number and chloroplast fine structure. Proc Natl Acad Sci 98(5):2473–2478. doi:10.1073/pnas.041620898

Hättenschwiler S, Körner C (1997) Biomass allocation and canopy development in spruce model ecosystems under elevated CO_2 and increased N deposition. Oecologia 113(1):104–114. doi:10.1007/s004420050358

Hutchison J, Manica A, Swetnam R, Balmford A, Spalding M (2014) Predicting global patterns in mangrove forest biomass. Conserv Lett 7(3):233–240. doi:10.1111/conl.12060

IPCC (2013) Climate change 2013: The physical science basis. Contribution of working group I to the fifth assessment report of the intergovernmental panel on climate change. Cambridge University Press, Cambridge, New York. doi:10.1017/CBO9781107415324

Jordan D, Ogren W (1984) The CO_2/O_2 specificity of ribulose 1,5-bisphosphate carboxylase/oxygenase. Planta 161(4):308–313. doi:10.1007/BF00398720

Kirschbaum MUF (2011) Does enhanced photosynthesis enhance growth? Lessons learned from CO_2 enrichment studies. Plant Physiol 155(1):117–124. doi:10.1104/pp.110.166819

Koch MS, Coronado C, Miller MW, Rudnick DT, Stabenau E, Halley RB, Sklar FH (2015) Climate change projected effects on coastal foundation communities of the Greater Everglades using a 2060 scenario: need for a new management paradigm. Environ Manag 55(4):857–875. doi:10.1007/s00267-014-0375-y

Körner C (2006) Plant CO_2 responses: an issue of definition, time and resource supply. New Phytol 172(3):393–411. doi:10.1111/j.1469-8137.2006.01886.x

Krause GH, Winter K, Krause B, Jahns P, García M, Aranda J, Virgo A (2010) High-temperature tolerance of a tropical tree, *Ficus insipida*: methodological reassessment and climate change considerations. Funct Plant Biol 37(9):890–900. doi:10.1071/FP10034

Krause GH, Winter K, Krause B, Virgo A (2014) Light-stimulated heat tolerance in leaves of two neotropical tree species, *Ficus insipida* and *Calophyllum longifolium*. Funct Plant Biol 42(1):42–51. doi:10.1071/FP14095

Krauss KW, McKee KL, Lovelock CE, Cahoon DR, Saintilan N, Reef R, Chen L (2014) How mangrove forests adjust to rising sea level. New Phytol 202(1):19–34. doi:10.1111/nph.12605

Leakey ADB, Xu F, Gillespie KM, McGrath JM, Ainsworth EA, Ort DR (2009) Genomic basis for stimulated respiration by plants growing under elevated carbon dioxide. Proc Natl Acad Sci 106(9):3597–3602. doi:10.1073/pnas.0810955106

Lloyd J, Farquhar GD (2008) Effects of rising temperatures and $[CO_2]$ on the physiology of tropical forest trees. Philos Trans R Soc Lond B Biol Sci. doi:10.1098/rstb.2007.0032

Long SP (1991) Modification of the response of photosynthetic productivity to rising temperature by atmospheric CO_2 concentrations: has its importance been underestimated? Plant Cell Environ 14(8):729–739. doi:10.1111/j.1365-3040.1991.tb01439.x

Luo Y, Su B, Currie WS, Dukes JS, Finzi A, Hartwig U, Hungate B, McMurtrie RE, Oren R, Parton WJ, Pataki DE, Shaw RM, Zak DR, Field CB (2004) Progressive nitrogen limitation of ecosystem responses to rising atmospheric carbon dioxide. Bioscience 54(8):731–739

McKee KL, Rooth JE (2008) Where temperate meets tropical: multifactorial effects of elevated CO_2, nitrogen enrichment, and competition on a mangrove-salt marsh community. Glob Change Biol 14(5):971–984. doi:10.1111/j.1365-2486.2008.01547.x

Miller PC (1972) Bioclimate, leaf temperature, and primary production in red mangrove canopies in South Florida. Ecology 53(1):22–45. doi:10.2307/1935708

Norby RJ, Warren JM, Iversen CM, Medlyn BE, McMurtrie RE (2010) CO_2 enhancement of forest productivity constrained by limited nitrogen availability. Proc Natl Acad Sci 107(45):19368–19373. doi:10.1073/pnas.1006463107

Nowak RS, Ellsworth DS, Smith SD (2004) Functional responses of plants to elevated atmospheric CO_2—do photosynthetic and productivity data from FACE experiments support early predictions? New Phytol 162(2):253–280. doi:10.1111/j.1469-8137.2004.01033.x

Oren R, Ellsworth DS, Johnsen KH, Phillips N, Ewers BE, Maier C, Schafer KVR, McCarthy H, Hendrey G, McNulty SG, Katul GG (2001) Soil fertility limits carbon sequestration by forest ecosystems in a CO_2-enriched atmosphere. Nature 411:469–472

Osland MJ, Enwright N, Day RH, Doyle TW (2013) Winter climate change and coastal wetland foundation species: salt marshes vs. mangrove forests in the southeastern United States. Glob Change Biol 19(5):1482–1494. doi:10.1111/gcb.12126

Phillips RP, Finzi AC, Bernhardt ES (2011) Enhanced root exudation induces microbial feedbacks to N cycling in a pine forest under long-term CO_2 fumigation. Ecol Lett 14(2):187–194. doi:10.1111/j.1461-0248.2010.01570.x

Pierret A, Gonkhamdee S, Jourdan C, Maeght J-L (2013) IJ_Rhizo: an open-source software to measure scanned images of root samples. Plant Soil 373(1–2):531–539. doi:10.1007/s11104-013-1795-9

Poorter H, Niklas KJ, Reich PB, Oleksyn J, Poot P, Mommer L (2012) Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. New Phytol 193(1):30–50. doi:10.1111/j.1469-8137.2011.03952.x

Quisthoudt K, Adams J, Rajkaran A, Dahdouh-Guebas F, Koedam N, Randin C (2013) Disentangling the effects of global climate and regional land-use change on the current and future distribution of mangroves in South Africa. Biodivers Conserv 22(6–7):1369–1390. doi:10.1007/s10531-013-0478-4

Reef R, Lovelock CE (2014a) Historical analysis of mangrove leaf traits throughout the 19th and 20th centuries reveals differential responses to increases in atmospheric CO_2. Glob Ecol Biogeogr 23(11):1209–1214. doi:10.1111/geb.12211

Reef R, Lovelock CE (2014b) Regulation of water balance in mangroves. Ann Bot. doi:10.1093/aob/mcu174

Reef R, Ball MC, Feller IC, Lovelock CE (2010a) Relationships among RNA: DNA ratio, growth and elemental stoichiometry in mangrove trees. Funct Ecol 24(5):1064–1072

Reef R, Feller IC, Lovelock CE (2010b) Nutrition of mangroves. Tree Physiol 30(9):1148–1160. doi:10.1093/treephys/tpq048

Reef R, Schmitz N, Rogers BA, Ball MC, Lovelock CE (2012) Differential responses of the mangrove *Avicennia marina* to salinity and abscisic acid. Funct Plant Biol 39(12):1038–1046. doi:10.1071/FP12178

Reef R, Winter K, Morales J, Adame MF, Reef DL, Lovelock CE (2015) The effect of atmospheric carbon dioxide concentrations on the performance of the mangrove *Avicennia germinans* over a range of salinities. Physiol Plant 154(3):358–368. doi:10.1111/ppl.12289

Reich PB, Hobbie SE, Lee T, Ellsworth DS, West JB, Tilman D, Knops JMH, Naeem S, Trost J (2006) Nitrogen limitation constrains sustainability of ecosystem response to CO_2. Nature 440(7086): 922–925. http://www.nature.com/nature/journal/v440/n7086/suppinfo/nature04486_S1.html

Saintilan N, Wilson NC, Rogers K, Rajkaran A, Krauss KW (2014) Mangrove expansion and salt marsh decline at mangrove poleward limits. Glob Change Biol 20(1):147–157. doi:10.1111/gcb.12341

Slot M, Winter K (2016) The effects of rising temperature on the ecophysiology of tropical forest trees. In: Goldstein G, Santiago LS (eds) Tropical tree physiology—adaptations and responses in a changing environment. Springer, Switzerland, p 467

Smith JAC, Popp M, Luttge U, Cram WJ, Diaz M, Griffiths H, Lee HSJ, Medina E, Schafer C, Stimmel KH, Thonke B (1989) Ecophysiology of xerophytic and halophytic vegetation of a coastal alluvial plain in northern Venezuela. VI. Water relations and gas exchange of mangroves. New Phytol 111(2):293–307. doi:10.2307/2556867

Taub DR, Seemann JR, Coleman JS (2000) Growth in elevated CO_2 protects photosynthesis against high-temperature damage. Plant Cell Environ 23:649–656

Team RDC (2014) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna

van der Sleen P, Groenendijk P, Vlam M, Anten NPR, Boom A, Bongers F, Pons TL, Terburg G, Zuidema PA (2015) No growth stimulation of tropical trees by 150 years of CO_2 fertilization but water-use efficiency increased. Nat Geosci 8(1):24–28. doi:10.1038/ngeo2313

Winter K, Aranda J, Garcia M, Virgio A, Paton S (2001a) Effect of elevated CO_2 and soil fertilization on whole-plant growth and water use in seedlings of a tropical pioneer tree, *Ficus insipida*. Flora Morphol Geobot Ecophysiol 196(6):458–464

Winter K, Garcia M, Gottsberger R, Popp M (2001b) Marked growth response of communities of two tropical tree species to elevated CO_2 when soil nutrient limitation is removed. Flora 196(1): 47–58

Woodroffe CD, Grindrod J (1991) Mangrove biogeography: the role of quaternary environmental and sea-level change. J Biogeogr 18(5):479–492. doi:10.2307/2845685

Yamori W, Hikosaka K, Way DA (2013) Temperature response of photosynthesis in C3, C4, and CAM plants: temperature acclimation and temperature adaptation. Photosynth Res 119(1):101–117. doi:10.1007/s11120-013-9874-6