

C₃ and C₄ Biomass Allocation Responses to Elevated CO₂ and Nitrogen: Contrasting Resource Capture Strategies

K. P. White · J. A. Langley · D. R. Cahoon ·
J. P. Megonigal

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Abstract Plants alter biomass allocation to optimize resource capture. Plant strategy for resource capture may have important implications in intertidal marshes, where soil nitrogen (N) levels and atmospheric carbon dioxide (CO₂) are changing. We conducted a factorial manipulation of atmospheric CO₂ (ambient and ambient+340 ppm) and soil N (ambient and ambient+25 gm⁻² year⁻¹) in an intertidal marsh composed of common North Atlantic C₃ and C₄ species. Estimation of C₃ stem turnover was used to adjust aboveground C₃ productivity, and fine root productivity was partitioned into C₃–C₄ functional groups by isotopic analysis. The results suggest that the plants follow resource capture theory. The C₃ species increased aboveground productivity under the added N and elevated CO₂ treatment ($P < 0.0001$), but did not under either added N or elevated CO₂ alone. C₃ fine root production decreased with added N ($P < 0.0001$), but fine roots increased under elevated CO₂ ($P =$

0.0481). The C₄ species increased growth under high N availability both above- and belowground, but that stimulation was diminished under elevated CO₂. The results suggest that the marsh vegetation allocates biomass according to resource capture at the individual plant level rather than for optimal ecosystem viability in regards to biomass influence over the processes that maintain soil surface elevation in equilibrium with sea level.

Keywords Biomass · Chesapeake Bay · Productivity · Sea level rise · Tidal marsh · Turnover

Introduction

Plants can optimize resource capture through shifts in biomass allocation under differing resource environments, and these shifts follow conserved patterns. Specifically, a tradeoff exists between producing aboveground structures for light capture and belowground structures for soil resource capture (Tilman and Wedin 1991; Craine 2009). Such patterns have been reported in ecosystems as diverse as grasslands (Suter et al. 2002) and forests (Pregitzer et al. 1995), and they tend to vary predictably across species according to photosynthetic functional group (Reynolds and Dantonio 1996).

The rules of resource capture and biomass allocation have especially important consequences in intertidal marsh ecosystems. Here, vegetation plays a direct role in sustaining the ecosystem. In tidal marshes, the presence of vegetation allows the ecosystem to maintain an equilibrium elevation relative to sea level (Redfield 1965; Morris 2006) by two primary mechanisms: (1) accumulation of organic soil by deposition of mostly endogenous organic matter (Turner 2004; Nyman et al. 2006; Mitsch and Gosselink 2007) and (2) trapping of exogenous sediments during tidal inundation (Morris et al.

K. P. White
Departments of Forest Ecology and Biogeosciences, and Statistical
Science, University of Idaho,
PO Box 83844-1133, Moscow, ID 83844, USA

J. A. Langley · J. P. Megonigal (✉)
Smithsonian Environmental Research Center,
PO Box 28, 647 Contees Wharf Road,
Edgewater, MD 21037, USA
e-mail: megonigalp@si.edu

J. A. Langley (✉)
Biology Department, Villanova University,
800 Lancaster Ave,
Villanova, PA 19085, USA
e-mail: adam.langley@villanova.edu

D. R. Cahoon
US Geological Survey, Patuxent Wildlife Research Center,
10300 Baltimore Avenue, BARC—East Building 308,
Beltsville, MD 20705, USA

2002; Mudd et al. 2010). Shifts in biomass allocation under an altered resource environment may alter these processes, and hence alter the viability of the marshes (Reed 1995; Morris et al. 2002).

Soil nitrogen (N) availability commonly limits marsh productivity (Valiela and Teal 1974) and is one of the more important resources affecting plant allocation. Under enhanced soil N availability, marsh plants may allocate more biomass aboveground, where aerobic decomposition decreases the amount of organic matter delivered to the soil. In turn, the shift in allocation aboveground may decrease the ability of the marsh to gain soil surface elevation relative to sea level where organic matter accumulation is most important to soil accumulation and hence marsh viability (Langley et al. 2009a). On the other hand, increases in atmospheric CO₂ often stimulate belowground production more than aboveground (Rogers et al. 1994; Iversen et al. 2008), but the stimulation usually depends on the photosynthetic functional group of the species (Poorter and Navas 2003). It remains unknown how large-scale perturbations such as rising CO₂ and nitrogen pollution will alter allocation patterns among marsh plants, which will have critical influence on plant-mediated mechanisms of soil elevation gain.

We conducted an experiment in a brackish tidal marsh to examine how plants alter plant biomass allocation patterns under combinations of enhanced soil N availability and increased atmospheric CO₂. To do this, we estimated biomass and productivity of C₃ and C₄ vegetation under factorial treatments of ambient and high soil N and atmospheric CO₂. Previous work reported earlier treatment effects on plot-level productivity (Langley et al. 2009a; Langley and Magonigal 2010); however, in this study, accounting for alteration in C₃ stem turnover allowed more accurate estimation of biomass allocation aboveground. Biomass allocation belowground is defined between the functional C₃–C₄ groups through carbon isotope analysis, allowing us to distinguish the strategies of these two functional groups that differ dramatically in photosynthetic strategy but commonly co-occur in brackish wetlands. Moreover, we take advantage of aboveground measurements of C₃ species in order to understand how changes in soil N availability may alter the ability of the marsh to trap sediments.

Site Description and Methods

Study Site

The study site is located in Kirkpatrick Marsh, the Smithsonian Institution's Global Change Research Wetland. Kirkpatrick Marsh is a high, brackish, intertidal marsh on the Chesapeake Bay in Maryland, in the USA (38°53' N, 76°33' W). The study plots were composed entirely of three species: a C₃ sedge,

Schoenoplectus americanus (formerly *Scirpus olneyi*), and two C₄ grasses, *Spartina patens* and *Distichlis spicata*. The soils of the marsh are histosols composed of more than 80 % organic matter. Active rooting by the marsh vegetation takes place in the upper 50 cm of the soil profile (Saunders et al. 2006). Directly below the active rooting zone, a more highly decomposed layer extends to a depth of 4–5 m where it meets a clayey mineral horizon. Salinity in the marsh is relatively low compared to other study systems, ranging from 4 to 15 ppt depending on regional rainfall. Tidal inundation of the marsh occurred in about 15 % of high ocean tides. More details of the site are given in Langley et al. (2009b).

Experimental Design

The open-top chambers used in this study were a modification of an earlier chamber design that had been rigorously tested for in situ elevated CO₂ experimentation (Drake et al. 1989). A chamber consisted of an octagonal frame and frustum, mounted on top of a hollow manifold for distributing air, which was mounted on an aluminum base that extended 30 cm into the soil. Each chamber enclosed 3.3 m² of marsh area. In this study, 20 of the chambers were installed in total. Detailed information about the chambers can be found in Langley et al. (2009a, b).

There were four experimental treatment groups consisting of atmospheric CO₂ and soil N additions in a 2×2 factorial design. Elevated atmospheric CO₂ was delivered in one of two levels, either ambient (i.e., no added atmospheric CO₂) or elevated CO₂ (ambient+340 ppm). Ten of the chambers received ambient CO₂. The other ten received the elevated CO₂ during daylight hours and ambient CO₂ during night. The CO₂ delivery occurred throughout each of two growing seasons, from late April through early November. The concentration of CO₂ inside each chamber was logged at 40-min intervals to document the precision of the elevated CO₂ treatments (Langley et al. 2009b). Like CO₂, N was added in one of two levels, either no enhancement, or at a rate of 25 g N m⁻² year⁻¹. The enhanced N level was applied to ten of the chambers in 5-g N m⁻² applications monthly from May through September each year. N was applied by spraying NH₄Cl (equivalent to 25 g N m⁻² year⁻¹) dissolved in local brackish water evenly across the plots. Following N application, an equal volume of unamended brackish water was sprayed onto the plots to rinse the N solution from the vegetation canopy to the soil surface. The enrichment in soil N availability was confirmed by elevated porewater [N] in the root zone, averaging increase over controls by 25 % over the course of the experiment (Langley and Magonigal 2010). The ten chambers that were to receive no N enhancement were sprayed with unamended brackish water in the same volume as the N enhanced chambers. The four treatments were (1) ambient CO₂ without N addition (Amb), (2) ambient CO₂

with N addition (Amb+N), (3) elevated CO₂ without N addition (Elev), and (4) elevated CO₂ with N addition (Elev+N). The experimental design was balanced ($n=5$ for each treatment).

Plant Growth Measurements

S. americanus aboveground biomass and productivity were estimated by a combination of annual harvest subsampling and tracking of stem turnover during the growing seasons. The harvests were undertaken in late July of each year by a method modified from Curtis et al. (1989). During the harvest, a subsample of *S. americanus* stems was measured, clipped, oven-dried at 60 °C for 72 h, and then weighed. The subsample measurements and weights were used to develop allometric equations ($r^2 > 0.9$ in both years) that were then used to estimate biomass during a larger, nondestructive sampling. The nondestructive sampling of *S. americanus* was conducted by measuring stem density, height, and width of shoots within six 30-cm² quadrats within each chamber. The area sampled totaled 16.3 % of the area within each chamber. The nondestructive measurements and allometric equations were used to estimate C₃ standing biomass for each season. We estimated C₃ shoot turnover by tracking a subsample of individually tagged *S. americanus* stems and monitoring the number of stems that senesced prior to harvest ($N=200$ in 2007 and $N=120$ in 2008). C₃ shoot productivity was then estimated as follows:

C₃ above ground productivity

$$= C_3 \text{ peak standing biomass} \\ + [(\text{fraction } C_3 \text{ dead at peak}) \times (C_3 \text{ peak standing biomass})]$$

where the fraction dead was the number of dead C₃ stems as a proportion of all stems (live + dead) over the course of the growing season.

Grass aboveground biomass (C₄ shoot biomass) was also estimated by measurements collected during the annual harvests; however, allometry was not used owing to the intractably large number of individual stems (Curtis et al. 1989). Instead, at each harvest, six 5-cm² areas were clipped from each chamber. The subsamples were oven-dried at 60 °C for 72 h and weighed, and then, those weights were scaled to the plot level as the estimates of C₄ shoot biomass. Though commonly used as a proxy for productivity, peak C₄ biomass underestimates productivity because shoot turnover and new growth after the peak biomass harvests were not measured.

Annual root productivity (C₃ and C₄ fine root, coarse roots, and rhizome) was estimated by weighing oven-dried, new root growth recovered from subsurface, mesh ingrowth bags. The bags were cylindrical, 5 cm in diameter,

and 30 cm in height. Each winter, in each chamber, three ingrowth bags were filled with root-free peat, inserted into precored holes, and retrieved the following winter. The functional group (C₃ or C₄) of coarse roots (>2 mm diameter) and rhizomes was determined by obvious visual characteristics (Saunders et al. 2006). For fine roots, visual distinction between the functional groups was not possible. Fine roots were separated into C₃ and C₄ functional groups using naturally occurring differences in $\delta^{13}\text{C}$ (Saunders et al. 2006). To measure the isotopic differentiation, the fine roots in the samples were ground and analyzed for ¹³C composition at the UC Davis Stable Isotope Laboratory using a PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). A mixing model was used to partition the fraction of fine root growth from *S. americanus* and the C₄ grasses as follows:

Fraction of fine root C from C₃ source

$$= (\delta^{13}\text{C}_{\text{fineroot}} - \delta^{13}\text{C}_{\text{C}_4} + \alpha) / (\delta^{13}\text{C}_{\text{C}_3} - \delta^{13}\text{C}_{\text{C}_4})$$

where $\delta^{13}\text{C}_{\text{fineroot}}$ is the ¹³C composition of a homogenized single fine root sample composited from the three replicate ingrowth bags from each plot. $\delta^{13}\text{C}_{\text{C}_3}$ and $\delta^{13}\text{C}_{\text{C}_4}$ represent foliar ¹³C for each functional group, determined separately for each plot to account for plot-level variability in strength of the novel ¹³C signature. An offset (α) was used to account for differences between shoots and roots arising either from fractionation or the incomplete integration of novel ¹³C into the root system (Langley et al. 2002). The α value was determined by comparing roots from a known species to shoots in the same plots for each CO₂ treatment group in each year (2007: ambient = -1.0‰, elevated = -0.5‰; 2008: ambient = -0.5‰, elevated = 1.1‰).

Trapping Index

To estimate how the treatments may alter the plant effect on sediment trapping of *S. americanus*, we calculated a sediment-trapping index for each of the treatments. The index is based on plant stem density (Leonard et al. 1995; Leonard and Luther 1995) and stem width and height (as suggested by the findings of Palmer et al. 2004). The index was calculated simply as the product of C₃ stem density, height, and width. Rather than predicting actual sediment accumulation at this sediment-poor site, this value was used to assess the potential for the global change treatments to affect sediment-trapping potential of one species.

Statistical Analyses

C₃ and C₄ standing biomass, C₃ shoot productivity, C₃ and C₄ coarse root and fine root productivity were analyzed separately. Because coarse roots and rhizomes showed

highly variable masses, rhizomes were included in “coarse roots” for analysis. We also analyzed aboveground plant parameters C_3 stem density, height and width, as well as the calculated trapping index. Before any modeling, univariate analysis on each of the analyzed response variables indicated normality (Shapiro–Wilks, $P>0.05$), so data transformation was unnecessary. Initially, the data were analyzed in MANOVAs with year as the within group variable. However, the year was generally insignificant, and at no point interacted with other factors, so the data over the 2 years were averaged for each plot. We followed by performing individual ANOVAs on each response. Fisher’s LSD multiple comparisons were conducted post hoc.

Results

C_3 aboveground biomass and productivity were both influenced by elevated CO_2 and enhanced N addition (Table 1). C_3 standing biomass decreased from the control when only N was added (Amb>Amb+N, $P=0.0423$), but when N was added in combination with elevated CO_2 , the C_3 standing biomass increased (Elev+N>Amb, $P=0.0034$; Fig. 1a). Accounting for turnover in the Amb+N and Elev+N treatments increased C_3 standing biomass estimates by 20–30 % (Fig. 1b; Table 2). Consequently, C_3 shoot productivity within treatments only differed from the control under the Elev+N treatment (Elev+N>Amb, $P<0.0001$; Fig. 1c).

For C_4 shoot biomass, CO_2 and N also interacted; however, the growth response pattern differed markedly from the C_3 species (Table 1; Fig. 2). C_4 shoot biomass was enhanced by N alone (Amb+N>Amb, $P<0.0001$). Under the Elev+N treatment, there was still an increase from the control (Elev+N–Elev>Amb, $P=0.0214$), but the gain was less than under the N only treatment (Amb+N>Elev+N, $P=0.0002$).

Belowground C_3 productivity was influenced differently by the treatments, depending on the particular root diameter class (Table 1). C_3 coarse root productivity did not change from the control under any of the treatments (Fig. 3a). There

Table 1 P values from two-way ANOVAs evaluating the treatment effects on C_3 and C_4 growth

		CO_2	N	$CO_2 \times N$
Standing biomass	C_3	<0.0001	0.9713	0.0057
Shoot productivity	C_3	<0.0001	0.0197	0.0106
	C_4	0.0026	<0.0001	0.0152
Coarse root productivity	C_3	0.4366	0.5137	0.5101
	C_4	0.1411	0.0161	0.0100
Fine root productivity	C_3	0.0481	<0.0001	0.6534
	C_4	0.5155	0.0363	0.0447

P values <0.05 are in bold

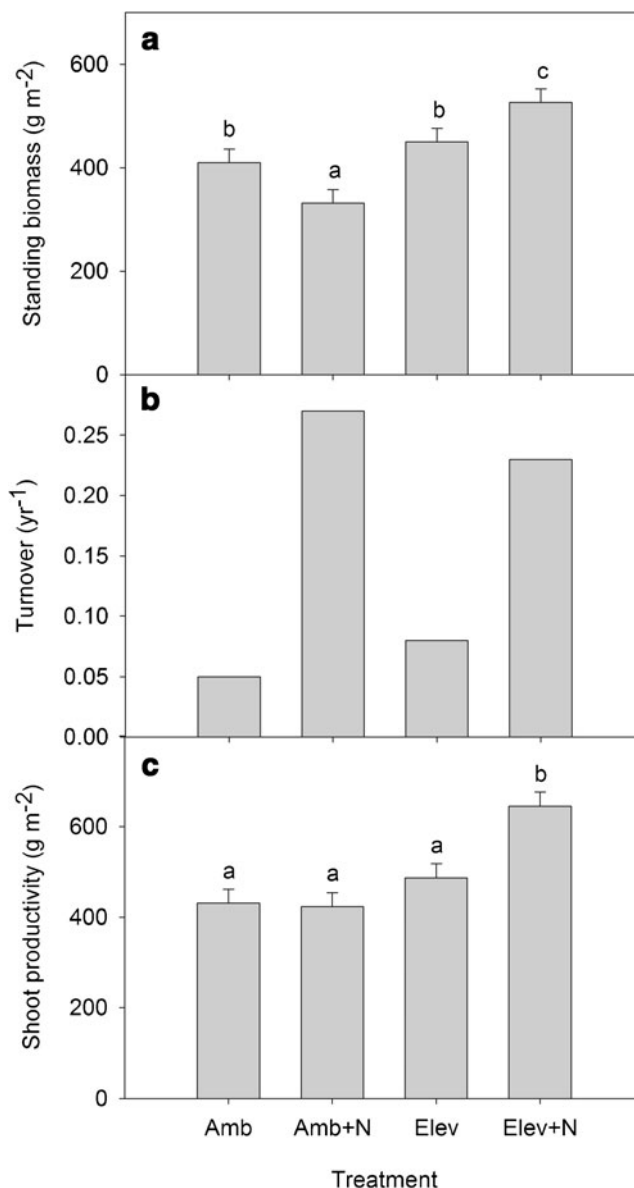


Fig. 1 C_3 **a** standing biomass, **b** stem turnover, and **c** shoot productivity. The four treatments were: (1) ambient CO_2 without N addition (Amb), (2) ambient CO_2 with N addition (Amb+N), (3) elevated CO_2 without N addition (Elev), and (4) elevated CO_2 with N addition (Elev+N). Columns represent means±standard error. ANOVA results are given in Table 1. Values with the same letter indicate no significant difference at $\alpha=0.05$

was no interaction between elevated CO_2 and N on fine root productivity, but the effects were additive. The fine root productivity decreased with added N (CO_2 treatments > N treatments, $P<0.0001$), but increased under elevated CO_2 (N treatments > CO_2 treatments, $P=0.0481$).

C_4 coarse root and fine root productivity responded similarly to the treatments (Table 1; Fig. 4). Both increased under added N without elevated CO_2 (Amb+N>Amb; coarse roots $P=0.0008$, fine roots $P=0.0053$). However, no other

Table 2 Adjustments made to the C_3 standing biomass to better estimate aboveground productivity

Treatment	Standing biomass (g m^{-2})	Turnover	Adjustment (g m^{-2})
Amb	409.6	0.05	+20.5
Amb+N	331.6	0.27	+89.5
Elev	449.6	0.08	+36.0
Elev+N	525.7	0.23	+120.9

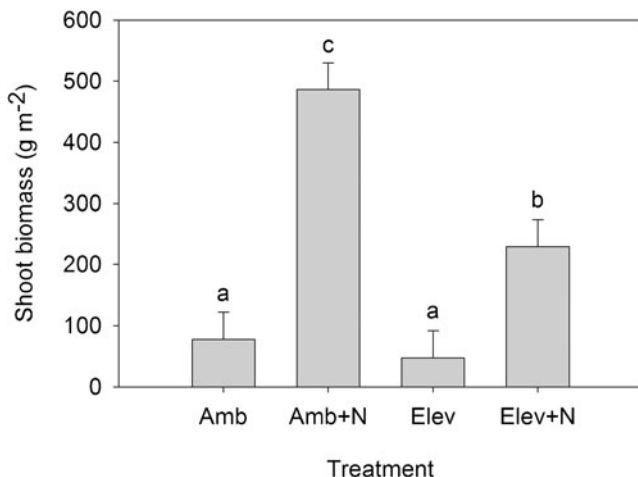
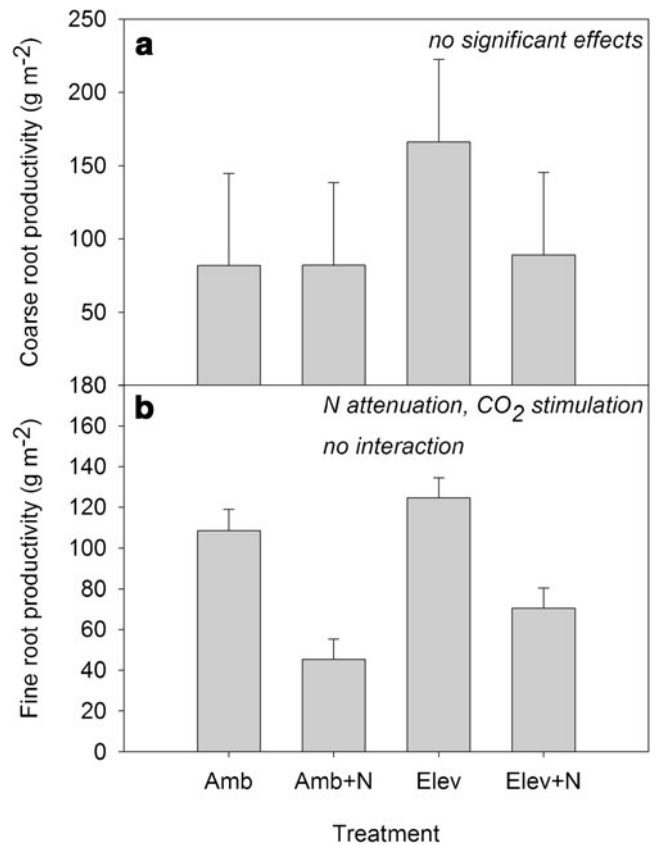
The four treatments were: (1) ambient CO_2 without N addition (Amb), (2) ambient CO_2 with N addition (Amb+N), (3) elevated CO_2 without N addition (Elev), and (4) elevated CO_2 with N addition (Elev+N)

treatments affected root productivity, indicating that elevated CO_2 reduced the N effect on C_4 belowground biomass.

The aboveground C_3 parameters that influence sediment trapping were each influenced by the treatments (Table 3). C_3 stem density only decreased under N alone (Amb+N < each of the other treatments, $P < 0.05$). In contrast, added N increased stem height and width (N treatments > CO_2 treatments; $P = 0.007$ C_3 stem height, $P = 0.0095$ C_3 stem width). The net effect on trapping was an increase only under both elevated CO_2 and added N (Elev+N > each of the other treatments, $P < 0.01$).

Discussion

Peak standing biomass has been commonly used as a proxy for treatment effects on aboveground productivity (Drake et al. 1989; Erickson et al. 2007). Nitrogen fertilization in other systems increases turnover of light-capturing organs in some species, in forests (Aerts 1989, 2009), and in

**Fig. 2** C_4 shoot biomass. The four treatments were: (1) ambient CO_2 without N addition (Amb), (2) ambient CO_2 with N addition (Amb+N), (3) elevated CO_2 without N addition (Elev), and (4) elevated CO_2 with N addition (Elev+N). Columns represent means \pm standard error. ANOVA results are given in Table 1. Values with the same letter indicate no significant difference at $\alpha = 0.05$ **Fig. 3** C_3 a coarse root growth and b fine root growth. The four treatments were: (1) ambient CO_2 without N addition (Amb), (2) ambient CO_2 with N addition (Amb+N), (3) elevated CO_2 without N addition (Elev), and (4) elevated CO_2 with N addition (Elev+N). Columns represent means \pm standard error. ANOVA results are given in Table 1

grasslands (Schlapfer and Ryser 1996). The present study corroborates those findings in the marsh. Here, we found that elevated CO_2 did not affect the turnover of individual *S. americanus* stems, while added N considerably accelerated turnover. Accounting for turnover where N was added, we adjusted the estimates, improving the accuracy of the aboveground allocation estimates in the sedge and altering the results for treatment effects on aboveground growth. The enhanced turnover effect of N was present even under elevated CO_2 conditions. As such, increased turnover due to N loading should be expected in the future when atmospheric CO_2 concentrations will be higher.

The stimulation in C_3 shoot productivity under the Elev+N treatment was not surprising in the context of resource capture. The increase in soil N availability under N additions would be expected to cause the sedge to shift allocation to organs for light capture (Tilman and Wedin 1991). The effect should be stronger under high CO_2 , since C_3 plants use atmospheric CO_2 inefficiently as compared to the C_4 plants (Bazzaz 1990). The alleviation of the belowground nutrient limitation and sparse atmospheric CO_2 constraint permit

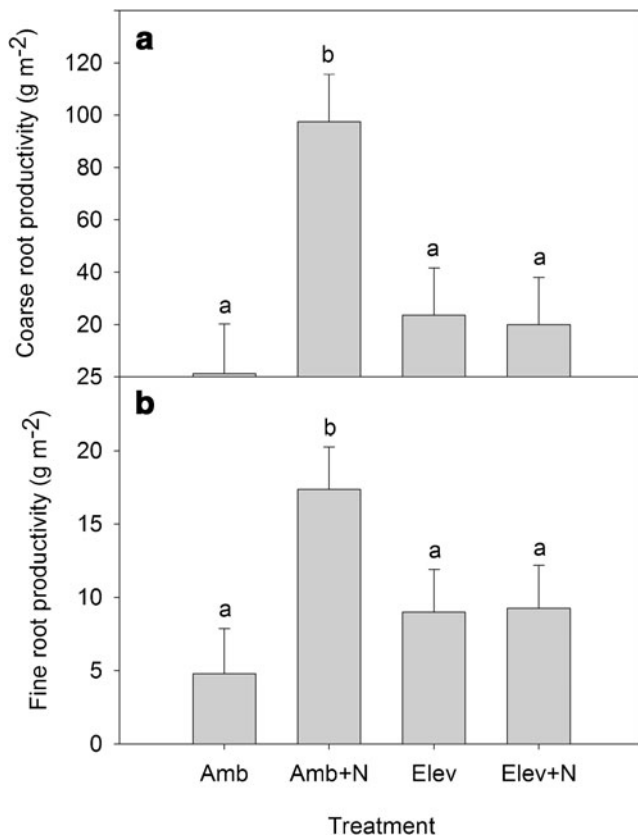


Fig. 4 *C*₄ **a** coarse root growth and **b** fine root growth. The four treatments were: (1) ambient CO₂ without N addition (*Amb*), (2) ambient CO₂ with N addition (*Amb+N*), (3) elevated CO₂ without N addition (*Elev*), and (4) elevated CO₂ with N addition (*Elev+N*). Columns represent means±standard error. ANOVA results are given in Table 1. Values with the same letter indicate no significant difference at $\alpha=0.05$

pursuit of light. Indeed, under the *Elev+N* treatment, shoots were taller and thicker than in the other treatments, suggesting that the shoots were acquiring more light (Carter and Knapp 2001). Moreover, the shoots in the *Elev+N* treatment were larger than under N addition without elevated CO₂, a change

that may indicate optimization related to CO₂ limitation (Ehleringer et al. 1991).

Belowground growth of *S. americanus* also followed the predicted resource capture patterns. Where N availability was enhanced, the sedge decreased allocation to roots in favor of new growth for light capture. The unresponsiveness of the *C*₃ coarse roots may have been due to high variability in the measurements. Nonetheless, the trend suggests that elevated CO₂ stimulates coarse roots if N scarcity constrains aboveground growth. The significant additive effects in the *C*₃ fine root productivity directly indicate that N addition decreases allocation belowground and CO₂ increases it. Moreover, other work suggests that as porewater N availability increases, belowground productivity in the system decreases (Langley and Megonigal 2010). It follows that the availability of soil N drives *C*₃ allocation belowground. Given the relative magnitude of belowground allocation between photosynthetic groups, the *C*₃ shift in allocation largely drives belowground allocation of the system. The effects on *C*₃ fine root productivity, excluding coarse roots, follow the responses one would predict if fine root growth is regulated by N capture (Pregitzer et al. 1995).

Most studies report that *C*₄ plant growth does not respond to elevated CO₂ (Ainsworth and Long 2005), however some suggest it does (Wand et al. 1999). In this study, the *C*₄ aboveground response contrasted with that of the *C*₃ species, and the lack of stimulation to CO₂ agrees with the biological theory. For the *C*₄ species, neither the linear dimensions on individual *C*₄ grass stems nor *C*₄ stem density was taken in this study, but it was visually apparent that enhancements at the individual *C*₄ plant level played a role in its biomass enhancement. The reduction in *C*₄ shoot biomass response under the *Elev+N* treatment relative to the added N alone was likely due to increased competition with the sedge for light capture (Emery et al. 2001), such that the diminished N response under CO₂ likely would not have occurred in a *C*₄ monoculture (Arp et al. 1993).

Table 3 Treatment means of parameters related to sediment-trapping properties for *S. americanus*

		Stem density (stems m ⁻²)	Stem height (cm)	Stem width (mm)	Trapping index (m ² m ⁻²)
Means	Amb	504 (35)	111.2 (1.2)	3.70 (0.05)	2.09 (0.2)
	Amb+N	423 (24)	116.8 (0.5)	3.79 (0.06)	1.87 (0.1)
	Elev	520 (23)	112.7 (2.4)	3.64 (0.06)	2.15 (0.2)
	Elev+N	581 (24)	121.3 (2.0)	3.91 (0.07)	2.76 (0.1)
Two-way ANOVA <i>P</i> values	CO ₂	0.005	0.096	0.664	0.006
	N	0.722	0.001	0.010	0.205
	CO ₂ × N	0.017	0.395	0.144	0.012

The four treatments were: (1) ambient CO₂ without N addition (*Amb*), (2) ambient CO₂ with N addition (*Amb+N*), (3) elevated CO₂ without N addition (*Elev*), and (4) elevated CO₂ with N addition (*Elev+N*). Standard errors are given in parentheses, and *P* values from a two-way ANOVA evaluating the effects of CO₂ and N on those parameters. *P* values <0.05 are in bold

Consequences for Marsh Elevation

Based on the relationships between biomass and sediment trapping established by previous research (e.g., Morris et al. 2002), our results suggest that N addition should generally enhance sediment trapping by increasing shoot biomass, while increasing CO₂ should enhance peat accumulation by increasing belowground productivity. In our system, further increases in N loading would appear to interrupt the peat formation from belowground productivity that sustains the high marsh.

Importantly, some of our findings may not be widely generalizable to the extent that species interactions vary from those observed in this marsh. For instance, the sediment-trapping capacity of the C₃ sedge in the ambient CO₂ + added N treatment is smaller than that in the ambient CO₂ only treatment. In this system the decline in C₄ biomass was driven by a negative interaction between the proliferating C₄ grasses and the C₃ sedge rather than a direct negative effect of the added N on the sedge. For that reason, we would not predict that either treatment should decrease aboveground growth parameters for either of the groups, C₃ or C₄ plants, in the absence of competition.

Other factors, such as global warming and sea level rise, may influence future marsh plant growth and allocation regionally. However, rising CO₂ concentrations are globally certain and relatively homogenous, and while patterns of N loading are more variable in space and time, N loading exerts a clear, robust control over growth allocation patterns. Importantly, N may directly stimulate microbial degradation of organic matter, which would also threaten marshes that depend on organic matter accumulation (Turner 2004), exacerbating attenuations in peat formation. Nutrient loading rates in the Chesapeake Bay are now much higher than in preindustrial times, but the levels have declined since the mid-1980s (Kemp et al. 2005). In light of the renewed commitment to decrease N and phosphorus loading into the Chesapeake Bay (Stokstad 2009), and evidence that regulation has reduced pollutant levels in a bay tributary (Ruhl and Rybicki 2010), forecasts of marsh responses to rising CO₂ should consider scenarios with both rising and falling trends in nutrient loading.

Conclusions

Taken together, the results of the study suggest that the marsh plants allocate biomass according to resource capture at the individual plant level, rather than allocating biomass for optimal ecosystem viability. The C₃ species allocates more belowground under low N availability, but more aboveground under high N. The C₄ species increase total biomass under high N availability, but the enhanced growth is likely due to

species expansion, rather than shifts at the individual level. The C₄ species are not directly responsive to CO₂. Differences in C₄ enhancement under elevated CO₂ to high N availability are likely mediated by interspecies competition. These responses suggest implications for the mechanisms that sustain marshes. Qualitatively, our results suggest that mineral-rich marshes may be able to trap more sediment with greater shoot growth under high N loading. However, N loading may reduce rates of soil accumulation in highly organic marshes, where the system depends on the deposition of organic matter belowground, not only by reducing allocation to roots in individual plants (as found by Turner 2011) but also by shifting community composition to species that innately produce less belowground matter.

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References

- Aerts, R. 1989. Above-ground biomass and nutrient dynamics of *Calluna vulgaris* and *Molinia caerulea* in a dry heathland. *Oikos* 56: 31–38.
- Aerts, R. 2009. Nitrogen supply effects on leaf dynamics and nutrient input into the soil of plant species in a sub-arctic tundra ecosystem. *Polar Biology* 32(2): 207–214.
- Ainsworth, E.A., and S.P. Long. 2005. What have we learned from 15 years of free-air CO₂ enrichment (FACE)? A meta-analytic review of the responses of photosynthesis, canopy. *New Phytologist* 165: 351–371.
- Arp, W.J., B.G. Drake, et al. 1993. Interactions between C-3 and C-4 salt-marsh plant-species during 4 years of exposure to elevated atmospheric CO₂. *Vegetatio* 104: 133–143.
- Bazzaz, F.A. 1990. The response of natural ecosystems to the rising global CO₂ levels. *Annual Review of Ecology and Systematics* 21: 167–196.
- Carter, G.A., and A.K. Knapp. 2001. Leaf optical properties in higher plants: Linking spectral characteristics to stress and chlorophyll concentration. *American Journal of Botany* 88: 677–684.
- Craine, J.M. 2009. *Resource strategies of wild plants*. Princeton: Princeton University Press.
- Curtis, P.S., B.G. Drake, et al. 1989. Growth and senescence in plant-communities exposed to elevated CO₂ concentrations on an estuarine marsh. *Oecologia* 78: 20–26.
- Drake, B.G., P.W. Leadley, et al. 1989. An open top chamber for field studies of elevated atmospheric CO₂ concentration on saltmarsh vegetation. *Functional Ecology* 3: 363–371.

- Ehleringer, J.R., R.F. Sage, et al. 1991. Climate change and the evolution of C_4 photosynthesis. *Trends in Ecology & Evolution* 6: 95–99.
- Emery, N.C., P.J. Ewanchuk, et al. 2001. Competition and salt-marsh plant zonation: Stress tolerators may be dominant competitors. *Ecology* 82: 2471–2485.
- Erickson, J.E., J.P. Megonigal, G. Peresta, and B.G. Drake. 2007. Salinity and sea level mediate elevated CO_2 effects on C_3 – C_4 plant interactions and tissue nitrogen in a Chesapeake Bay tidal wetland. *Global Change Biology* 13(1): 202–215.
- Iversen, C.M., J. Ledford, and R.J. Norby. 2008. CO_2 enrichment increases carbon and nitrogen input from fine roots in a deciduous forest. *New Phytologist* 179: 837–847.
- Kemp, W.M., W.R. Boynton, et al. 2005. Eutrophication of Chesapeake Bay: Historical trends and ecological interactions. *Marine Ecology Progress Series* 303: 1–29.
- Langley, J.A., and J.P. Megonigal. 2010. Ecosystem response to elevated CO_2 levels limited by nitrogen-induced plant species shift. *Nature* 466: 96–99.
- Langley, J.A., B.G. Drake, et al. 2002. Extensive belowground carbon storage supports roots and mycorrhizae in regenerating scrub oaks. *Oecologia* 131: 542–548.
- Langley, J.A., K.L. McKee, et al. 2009a. Elevated CO_2 stimulates marsh elevation gain, counterbalancing sea-level rise. *Proceedings of the National Academy of Sciences of the United States of America* 106: 6182–6186.
- Langley, J.A., M.V. Sigrist, J. Duls, D.R. Cahoon, J.C. Lynch, and J.P. Megonigal. 2009b. Global change and marsh elevation dynamics: Experimenting where land meets sea and biology meets geology. *Smithsonian Contributions to the Marine Sciences* 38: 391–400.
- Leonard, L.A., and M.E. Luther. 1995. Flow hydrodynamics in tidal marsh canopies. *Limnology and Oceanography* 40: 1474–1484.
- Leonard, L.A., A.C. Hine, et al. 1995. Surficial sediment transport and deposition processes in a *Juncus roemerianus* marsh, west-central Florida. *Journal of Coastal Research* 11: 322–336.
- Mitsch, W.J., and J.G. Gosselink. 2007. *Wetlands*. Hoboken: Wiley.
- Morris, J.T. 2006. Competition among marsh macrophytes by means of geomorphological displacement in the intertidal zone. *Estuarine, Coastal and Shelf Science* 69: 395–402.
- Morris, J.T., P.V. Sundareshwar, et al. 2002. Responses of coastal wetlands to rising sea level. *Ecology* 83: 2869–2877.
- Mudd, S.M., A. D'Alpaos, and J.T. Morris. 2010. How does vegetation affect sedimentation on tidal marshes? Investigating particle capture and hydrodynamic controls on biologically mediated sedimentation. *Journal of Geophysical Research—Earth Surface* 115: f3.
- Nyman, J.A., R.J. Walters, et al. 2006. Marsh vertical accretion via vegetative growth. *Estuarine, Coastal and Shelf Science* 69: 370–380.
- Palmer, M.R., H.M. Nepf, et al. 2004. Observations of particle capture on a cylindrical collector: Implications for particle accumulation and removal in aquatic systems. *Limnology and Oceanography* 49: 76–85.
- Poorter, H., and M.L. Navas. 2003. Plant growth and competition at elevated CO_2 : On winners, losers and functional groups. *New Phytologist* 157: 175–198.
- Pregitzer, K.S., D.R. Zak, P.S. Curtis, M.E. Kubiske, J.A. Teeri, and C.S. Vogel. 1995. Atmospheric CO_2 , soil-nitrogen and turnover of fine roots. *New Phytologist* 129(4): 579–585.
- Redfield, A.C. 1965. Ontogeny of a salt marsh estuary. *Science* 147: 50–55.
- Reed, D.J. 1995. Sediment dynamics, deposition and erosion in temperate salt marshes. *Journal of Coastal Research* 11: 295–295.
- Reynolds, H.L., and C. Dantonio. 1996. The ecological significance of plasticity in root weight ratio in response to nitrogen: Opinion. *Plant and Soil* 185: 75–97.
- Rogers, H.H., G.B. Runion, and S.V. Krupa. 1994. Plant-responses to atmospheric CO_2 enrichment with emphasis on roots and the rhizosphere. *Environmental Pollution* 83: 155–189.
- Ruhl, H.A., and N.B. Rybicki. 2010. Long-term reductions in anthropogenic nutrients link to improvements in Chesapeake Bay habitat. *Proceedings of the National Academy of Sciences of the United States of America* 107: 16566–16570.
- Saunders, C.J., J.P. Megonigal, et al. 2006. Comparison of belowground biomass in C-3- and C-4-dominated mixed communities in a Chesapeake Bay brackish marsh. *Plant and Soil* 280: 305–322.
- Schlapfer, B., and P. Ryser. 1996. Leaf and root turnover of three ecologically contrasting grass species in relation to their performance along a productivity gradient. *Oikos* 75: 398–406.
- Stokstad, E. 2009. Obama moves to revitalize Chesapeake Bay restoration. *Science* 324(5931): 1138–1139.
- Suter, D., M. Frehner, et al. 2002. Elevated CO_2 increases carbon allocation to the roots of *Lolium perenne* under free-air CO_2 enrichment but not in a controlled environment. *New Phytologist* 154: 65–75.
- Tilman, D., and D. Wedin. 1991. Plant traits and resource reduction for 5 grasses growing on a nitrogen gradient. *Ecology* 72: 685–700.
- Turner, R.E. 2004. Coastal wetland subsidence arising from local hydrologic manipulations. *Estuaries* 27: 265–272.
- Turner, R.E. 2011. Beneath the salt marsh canopy: Loss of soil strength with increasing nutrient loads. *Estuaries and Coasts* 34: 1084–1093.
- Valiela, I., and J.M. Teal. 1974. Nutrient limitation in salt marsh vegetation. In *Ecology of halophytes*, ed. R.J. Reimold and W.H. Queen, 574. New York: Academic Press.
- Ward, S.J.E., G.F. Midgley, et al. 1999. Responses of wild C_4 and C_3 grass (Poaceae) species to elevated atmospheric CO_2 concentration: A meta-analytic test of current theories and perceptions. *Global Change Biology* 5: 723–741.