Global Change Biology

Global Change Biology (2013) **19**, 2976–2985, doi: 10.1111/gcb.12287

Drought-induced saltwater incursion leads to increased wetland nitrogen export

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Abstract

Coastal wetlands have the capacity to retain and denitrify large quantities of reactive nitrogen (N), making them important in attenuating increased anthropogenic N flux to coastal ecosystems. The ability of coastal wetlands to retain and transform N is being reduced by wetland losses resulting from land development. Nitrogen retention in coastal wetlands is further threatened by the increasing frequency and spatial extent of saltwater inundation in historically freshwater ecosystems, due to the combined effects of dredging, declining river discharge to coastal areas due to human water use, increased drought frequency, and accelerating sea-level rise. Because saltwater incursion may affect N cycling through multiple mechanisms, the impacts of salinization on coastal freshwater wetland N retention and transformation are not well understood. Here, we show that repeated annual saltwater incursion during late summer droughts in the coastal plain of North Carolina changed N export from organic to inorganic forms and led to a doubling of annual NH_4^+ export from a 440 hectare former agricultural field undergoing wetland restoration. Soil solution NH_4^+ concentrations in two mature wetlands also increased with salinization, but the magnitude of increase was smaller than that in the former agricultural field. Long-term saltwater exposure experiments with intact soil columns demonstrated that much of the increase in reactive N released could be explained by exchange of salt cations with sediment NH_4^+. Using these findings together with the predicted flooding of 1661 km^2 of wetlands along the NC coast by 2100, we estimate that saltwater incursion into these coastal areas could release up to 18 077 Mg N, or approximately half the annual NH_4^+ flux of the Mississippi River. Our results suggest that saltwater incursion into coastal freshwater wetlands globally could lead to increased N loading to sensitive coastal waters.

Keywords: cation exchange, eutrophication, land-use change, salinization, sea-level rise

Received 25 February 2013 and accepted 6 May 2013

Introduction

In low-lying coastal plains throughout the world, extensive freshwater marshes, swamps, and tidal creeks are connected via surface water to the coastal ocean. In these systems, extended drought or wind surges can increase the upstream reach of brackish water and the peak salinity of high tides. This saltwater incursion, or landward movement of salinity from the ocean onto the coastal plain, represents the leading edge of climate change for coastal plain ecosystems (Sklar & Browder, 1998; Day *et al.*, 2007; Anderson & Lockaby, 2011). In the southeastern US, most climate change models suggest that both the duration and severity of droughts and the frequency and intensity of hurricane storms will increase (Mulholland *et al.*, 1997), facilitating both seasonal and episodic saltwater incursion. Human-accelerated sea-level rise exacerbates this increased potential for the chemical diffusion and mixing of salts from sea to land. Saltwater incursion into freshwater areas leads to osmotic stress for vascular plants, increases in tree mortality, and over time eventually leads to the conversion from forested wetlands to oligohaline or brackish marshes (Brinson *et al.*, 1995; Day *et al.*, 2007; Krauss *et al.*, 2009; Craft, 2012).

Freshwater wetlands are important sinks for reactive nitrogen (N, Jordan *et al.*, 2011). However, as freshwater wetlands become brackish, their capacity to retain or denitrify N may substantially decline (Gardner *et al.*, 1991; Seitzinger *et al.*, 1991; Craft *et al.*, 2009; Chambers *et al.*, 2011). Vascular plant N uptake rates may slow as sulfide (HS^-) toxicity and increased salt stress decrease plant growth (Cormier *et al.*, 2013). Ammonium ions (NH_4^+) sorbed on cation exchange sites in wetland soils can be displaced by the influx of sea salt cations (Na^+, Mg^{2+}, Ca^{2+}, Gardner *et al.*, 1991; Giblin *et al.*, 2010; Seitzinger *et al.*, 1991; Weston *et al.*, 2010). Simultaneously, increases in sulfate (SO_4^{2-}) concentrations will likely enhance rates of organic matter mineralization in waterlogged sediments via SO_4^{2-} reduction to sulfide (HS^-), leading to the release of organically bound N as

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NH_4^+(Caraco *et al.*, 1989; Lamers *et al.*, 1998). Other microbial responses to saltwater are not as well understood, but there is evidence to suggest that high concentrations of sea salts may inhibit rates of nitrification ($NH_4^+ \rightarrow NO_3^-$, Joye & Hollibaugh, 1995) while enhancing rates of dissimilatory nitrate (NO_3^-) reduction to NH_4^+ (Gardner *et al.*, 2006), two mechanisms that would each lead to increases in dissolved NH_4^+ concentrations. Resulting reductions in the supply of NO_3^- through decreased nitrification together with the potential toxicity of HS^- to denitrifiers could each reduce rates of denitrification ($NO_3^- \rightarrow N_2$, Rysgaard *et al.*, 1999). Given that these mechanisms could occur at different temporal and spatial scales, and that most of this research has been conducted on soil core or m^2 plot scales manipulations of the sediment/water interface, we lack a clear understanding of the consequences of saltwater incursion on N cycling at the ecosystem scale.

Interactions between saltwater incursion and land use (both past and current) make it difficult to predict the response of coastal plain wetlands to climate change. In the southeast US and Europe, much of the coastal plain wetlands were drained or filled for agriculture (Barendregt *et al.*, 2009). In the North Carolina coastal plain, colonial-era drainage networks were vastly expanded in the 1970s and 1980s, allowing the conversion of much of the region to intensive row crop agriculture and consolidated animal feeding operations (Carter, 1975; Dahl, 1990). While losses in soil elevation of up to 1 m via oxidation of soil organic matter in cultivated soils (O'Driscoll, 2012) have increased the areas susceptible to saltwater incursion, active pumping and water control structures tend to artificially decrease the current areal extent of saltwater incursion in actively managed agricultural lands. Furthermore, both current and past widespread inorganic fertilizer application increase the potential for large-scale salt-mediated losses of legacy nutrients from these soils. A more complete understanding of how wetlands with different land-use histories may respond to increased salinization will be necessary to forecast the response of these sensitive coastal landscapes to climate change.

Here, we synthesize 5 years of water and nutrient exports from the Timberlake Observatory for Wetland Restoration (TOWeR), soil solution nutrient concentrations from two mature forested wetlands, and a mechanistic 12-week laboratory microcosm study to examine the impacts of saltwater incursion on N export and cycling. We previously reported high rates of annual NO_3^- retention accompanied by release of NH_4^+ and dissolved organic N from the TOWeR site (Ardón *et al.*, 2010a). We hypothesized that saltwater incursion would increase the release and export of reactive forms of N in all sites and in the microcosm experiment, but that the magnitude would be greater in the restored wetland (due to its past agricultural use) than in the mature forested wetlands.

Materials and methods

Site description

This study was conducted in the Timberlake Observatory for Wetland Restoration (TOWeR), located in the Albemarle Peninsula in Tyrell Count, North Carolina (35°54′22″ N 76°09′25″ W, Fig. 1). The site is part of the Great Dismal Swamp Mitigation Bank, LLC, a 1704.2 ha compensatory mitigation bank composed of: 420 ha of mature forested wetland, 787 ha of forested wetland, 57.2 ha of drained shrub-scrub, and 440 ha of former agricultural fields undergoing stream and wetland restoration. The elevation across the site ranges from −1 to 2 m above sea level (lidar survey by National Center for Airborne Laser Mapping 2008, Houston, Texas, USA). Surface water leaving the site flows to the Albemarle Sound via the

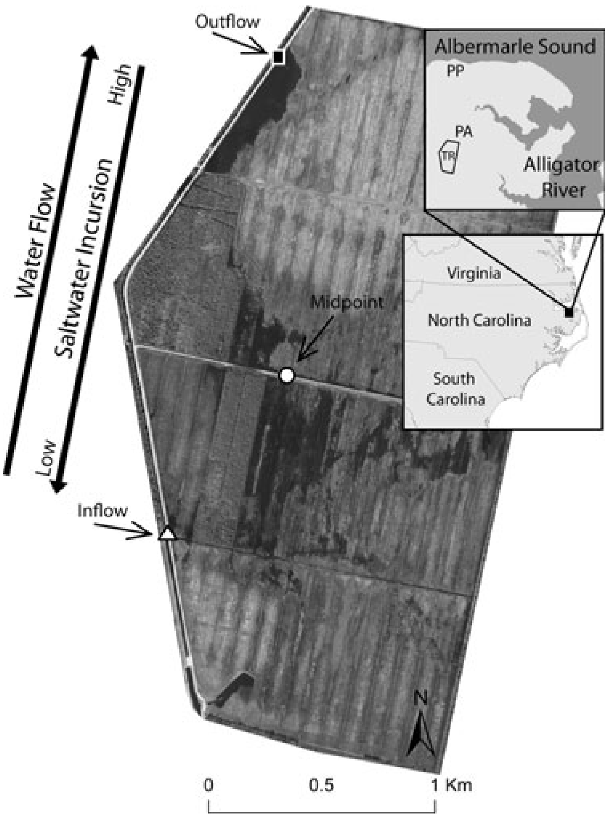

Fig. 1 Location of sampling sites in North Carolina (inset, PP, Palmetto Peartree Preserve; PA, Preservation area; TR, Timberlake Observatory for Wetland Restoration). Map of Timberlake Observatory for Wetland Restoration (TOWeR) with location of surface water sampling sites. Arrows indicate direction of predominant water flow (downstream) and saltwater incursion (upstream).

Little Alligator River. A detailed description of the history of the site and restoration practices has been published elsewhere (Ardón *et al*., 2010a). The Albemarle Sound is a microtidal system with wind-driven tides and brackish salinity [0–7 parts per thousand (ppt), Corbett *et al*., 2007]. During drought years, salinities of up to 12 ppt have been measured in the Albemarle Sound (unpublished data, Division of Marine Fisheries, North Carolina Department of Environment and Natural Resources).

To examine the response to increased salinity of wetlands without past agricultural land use, we also sampled soil solution nutrient concentrations from two established mature forested wetlands. The first site is a preservation area (PA) located within a minimally impacted stand within the TOWeR site, dominated by mixed hardwood forest (oak-tupelo-cypress, Fig. 1, Ardón *et al*., 2010b; Morse *et al*., 2012). The other established forested wetland site is in the Palmetto Peartree Preserve (PP), in a swamp hardwood stand of cypress and tupelo (Ardón *et al*., 2010b; Morse *et al*., 2012), located 8 km away from TOWeR and adjacent to the Albemarle Sound (Fig. 1).

Surface water sampling

Starting in February 2007, two water samples were collected weekly from three sampling locations along the site: Inflow, Midpoint, and Outflow (Fig. 1). One water sample was filtered in the field (GF/F Whatman filters, 0.7 μm) and the other was left unfiltered for total nutrient digestions. Samples were refrigerated and maintained at 4 °C until analyzed (within 3 weeks of collection). NH_4-N was measured using the phenate method on a Lachat QuickChem automated system. NO_3-N, Cl^-, and SO_4^{2-} were measured on a Dionex ICS-2000 ion chromatograph (Dionex Corporation, Sunnyvale, CA, USA). Total dissolved nitrogen (TDN) was measured on a Shimadzu TOC-V total carbon analyzer with a TNM-1 nitrogen module (Shimadzu Scientific Instruments, Columbia, Maryland, USA). We calculated dissolved organic nitrogen (DON) concentrations as DON = TDN - (NO_3-N + NH_4-N). Water conductivity, pH, dissolved oxygen, and temperature were measured using a handheld device (YSI Multiprobe Model 560; Yellow Springs, OH, USA), at the same time the water samples were collected. We used measured Cl^- concentrations to estimate salinity (in parts per thousand).

Soil solution sampling

To examine solutes in soil solution we sampled a network of piezometer and lysimeters across 33 sampling stations in the TOWeR site every 2–4 months. Near-surface samples (15 cm depth) were collected from piezometers installed in the wetter areas and from lysimeters in infrequently flooded areas. In seven sites, we also installed piezometers at 30, 60, and 100 cm. We purged the piezometers or applied a vacuum (40 kPa) to the lysimeters, and then collected the samples the next day. Samples were collected in high density polyethylene plastic (HDPE) plastic bottles, one unfiltered and one filtered (Whatman GF/F. 0.7 μm) and refrigerated until analyzed as described above. Piezometers and lysimeters were also installed in five sampling stations in each of the two established wetland sites.

Water and nutrient export

At the TOWeR site, water depth and velocity have been measured in the single constrained outflow (Outflow) from 25 February 2007 to 25 February 2012. Water depth and velocity were measured every 15 min using acoustic Doppler area velocity meters (2150 Area Velocity Meter, Teledyne ISCO, Lincoln, NE, USA). We used the known area of the culvert, water depth, and velocity to estimate discharge. Bidirectional flow and slow velocity made measurements difficult and at times unreliable. We excluded outliers (more than one order of magnitude greater than the highest velocity recorded using a handheld flow meter) and interpolated between data points using weighted averages (Ardón *et al*., 2010a). Rainfall was recorded using tipping bucket gauges in three locations throughout the site (0.01 inch rain tip gauge, Teledyne ISCO). To estimate nutrient fluxes out of the site we used the concentrations measured in our weekly sampling and daily discharge. For dates between samples we used the average concentration from the beginning and ending values multiplied by discharge during the time period (Likens & Bormann, 1995). We were unable to estimate nutrient exports for the established wetlands due to the long residence time and slow flows in drainage canals surrounding the sites.

Microcosm experiment

To examine the mechanisms driving changes in N release, we conducted a microcosm experiment using intact soil cores from the TOWeR site. We designed the experiment to separate the effects of drought from saltwater incursion and to isolate the role of increased SO_4^{2-} on NH_4^+ release and nitrification potential. The design allowed us to distinguish between changes due to salinity through cation exchange and changes in microbial activity associated with increased SO_4^{2-} concentrations. We collected intact soil cores (30 cm deep, 2.5 cm diameter) to build microcosms based on a design modified from Clark *et al*. (2006). The microcosms consist of the main soil core connected to a reservoir (30 cm tall, 5 cm diameter, PVC) that allowed us to manipulate both the chemistry of soil solution and the water elevation within the soil core (Fig. S1). Each core was instrumented with three microlysimeters (5 cm, glass fiber, Rhizon MOM, Rhizosphere Research Products, the Netherlands) at three depths (−5, −15 and −25 cm) to sample soil solution. On 26 July 2010, we collected 50 soil cores from a site within TOWeR that has not experienced saltwater incursion during our sampling period. Ten soil cores were used to determine initial soil nutrient and physical characteristics. The other 40 cores were set up in a 2 × 2 × 2 full factorial design with hydrology, salt, and sulfate as factors. We modified a known recipe for artificial saltwater (ASW) (5 ppt salinity, Kester *et al*., 1967) to make the following chemical treatments: control (deionized water), ASW, artificial saltwater without sulfate (ASW-SO_4), and deionized water with the same SO_4^{2-} concentrations as the ASW treatments (SO_4). The cores were incubated in a dark environmental chamber at 30 °C and 20% humidity for the 12 weeks of the experiment. We maintained the cores at 30 °C because this temperature is typical of late summer at our field site (mean daily temperature July–September is 25.6 °C, with the maximum measured 46.7 °C), when salinity tends to occur. The cores were set up in the chamber on 9 August 2010, flooded with deionized water, and allowed to equilibrate for 2 weeks before applying the treatments. In the flooded treatments, the water level was maintained at the soil surface, and for the drought treatment at the -25 cm sampling microlysimeter for the duration of the experiment. Water elevations in the reservoirs were monitored every 2 days, refilling with the appropriate solution when necessary. Refilling the reservoirs with treatment solution led to incremental increases in salinity over time, as has been documented in the field during drought periods.

We sampled soil solution on 24 August 2010, before implementing the treatments. The chemical and drought treatments were implemented on 25 August 2010. We sampled soil solution chemistry weekly for the first 4 weeks after treatments were initiated, then switched to biweekly sampling for the remainder of the experiment. Soil solution was sampled by connecting a 5 ml syringe to each of the microlysimeters, adding a vacuum and leaving the syringes to collect soil solution for 12 h (Clark *et al.*, 2006). We collected samples in a clean HDPE scintillation vials, and samples were either analyzed immediately or frozen until analysis (within 3 weeks). Soil solution samples were analyzed for TDN, Cl^-, SO_4^{2-}, NO_3-N, and NH_4-N as described above. Soils were sampled at the end of the experiment to measure nitrification potential using methods described in Fierer & Schimel (2002).

Statistical analyses

To examine the effects of saltwater incursion on N concentrations within each site, we compared N concentrations from samples that had salinities below the brackish limit of 0.5 ppt to N concentrations from samples with salinities above the brackish limit using *t*-tests. Analysis of variance was used on the microcosm ammonium and nitrification potential data using hydrology, salinity, and sulfate as factors followed by a post hoc Tukey test of means. Data were log transformed to meet assumptions of normality.

Results

During four of the five sampling years TOWeR experienced drought conditions starting in late summer, leading to saltwater incursion through surface water (Fig. 2). Salinity in surface water at the site is usually <0.5 ppt, but during saltwater incursion events it increased to >5 ppt (Fig. 2). Soil solution Cl^- concentrations within TOWeR site decreased with depth, confirming that saltwater incursion occurred via surface water (Table 1). Cl^- concentrations did not change with depth at PP, due to its close proximity to the Albemarle Sound (Table 1, Fig. 1). Peak Cl^- concentrations were lower in the PA site (a hydrologically isolated wetland, <100 mg l^{-1} or <0.5 ppt salinity) compared to TOWeR (the restored site) and PP (the sound-side wetland) sites where Cl^- reached as high as 1500 mg l^{-1} (or 2.7 ppt salinity, Table 1, Fig. 3). In all three wetlands, ammonium concentrations increased with increasing Cl^- concentrations (Fig. 3).

Drought-induced saltwater incursion at TOWeR was associated with higher NH_4^+ concentrations in surface water (fourfold) and soil solution (sevenfold, Fig. 3a and b, $P < 0.001$ for both). At similar salinities, NH_4^+ concentrations in surface water were often higher during the non growing season (November to February) than during the growing season (March through October, Fig. 3b). There were six different time periods that experienced high Cl^- but not high NH_4^+ during the growing season (Fig. 3b); this was in part because increases in NH_4^+ concentrations in surface water often lagged increases in Cl^- concentrations by 1–2 months (Fig. S2). During saltwater incursion events, the Cl^- to SO_4^2 ratio increased as saltwater moved upstream across the different sites (Fig. S3). Saltwater incursion also changed the dominant form of N export from primarily organic (DON) to inorganic (Table 2), and led to a twofold to fourfold increase in annual NH_4^+ export during years of high water salinity (Table 2).

Microcosm experiment

Chloride concentrations reached more than 2000 mg l^{-1} in the two saltwater treatments (ASW with and without SO_4^{2-}) as compared to <20 mg l^{-1} in the control or SO_4^{2-} only treatment (Fig. 4a–d). Sulfate concentrations reached more than 100 mg l^{-1} in the SO_4^{2-} only treatments and the ASW with SO_4^{2-} as compared to less than 30 mg l^{-1} in the control and ASW without SO_4^{2-} (Fig. 4e–h). The experimental drought lowered NH_4^+ concentrations in the control and SO_4^{2-} treatments, but there was no difference in the two salt treatments (Fig. 4i–l, significant effect hydrology and hydrology*salt interactions, Table 3). The addition of salt (both with and without SO_4^{2-}) increased NH_4^+ release, leading to fivefold to tenfold more NH_4^+ than either the control or SO_4^{2-} only treatments (Fig. 4, significant effect of salt, Table 3). Nitrification potential responded more strongly to hydrologic treatments; with rates being 20 times higher in all the drought treatments than in all the flooded treatments (Fig. 5, significant hydrology effect, Table 3). There were no significant effects of salt or sulfate on nitrification potential in the flooded cores, but there was a significant interaction between hydrology and SO_4^{2-} treat-

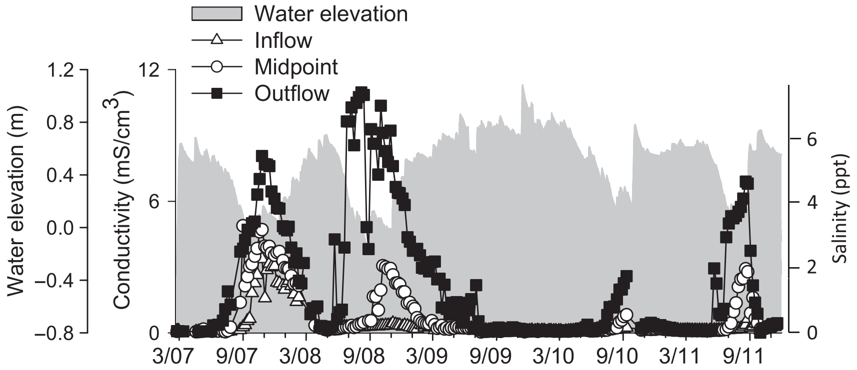

Fig. 2 Water surface elevation (gray area, in m above datum NGVD 29), water conductivity (mS cm^{-3}) and salinity (parts per thousand, right *y*-axis) in three sites across the Timberlake Observatory for Wetland Restoration (TOWeR).

Table 1 Average soil solution solute concentrations from three wetland sites

	NH_4-N (mg l^{-1})	NO_3-N (mg l^{-1})	DON (mg l^{-1})	TDN (mg l^{-1})	Cl^- (mg l^{-1})	SO_4^{2-} (mg l^{-1})
TOWeR						
15 cm	5.26 ± 7.09	0.06 ± 0.10	2.74 ± 2.64	8.28 ± 8.38	457.47 ± 454.7	18.00 ± 21.9
30 cm	4.47 ± 10.4	0.04 ± 0.07	1.51 ± 1.19	6.01 ± 11.2	213.30 ± 289.0	15.76 ± 11.8
60 cm	1.20 ± 2.67	0.06 ± 0.09	1.02 ± 0.78	2.29 ± 2.95	166.67 ± 215.8	23.31 ± 16.4
100 cm	0.45 ± 1.19	0.03 ± 0.03	0.98 ± 0.92	1.47 ± 1.82	73.00 ± 73.9	27.58 ± 28.8
PA						
15 cm	0.30 ± 0.28	0.04 ± 0.02	0.79 ± 0.43	1.15 ± 0.55	42.00 ± 24.8	31.50 ± 20.6
30 cm	0.34 ± 0.19	0.13 ± 0.21	1.32 ± 1.37	1.79 ± 1.53	33.00 ± 9.83	34.20 ± 17.9
60 cm	0.07 ± 0.05	0.03 ± 0.01	1.48 ± 0.81	1.58 ± 0.83	103.00 ± 46.7	96.22 ± 39.7
100 cm	0.10 ± 0.04	0.02 ± 0.02	2.00 ± 0.45	2.13 ± 0.46	139.00 ± 60.7	125.08 ± 23.0
PP						
15 cm	0.73 ± 0.61	0.03 ± 0.03	0.86 ± 0.89	1.45 ± 0.64	1859.0 ± 895.4	167.80 ± 99.1
30 cm	0.68 ± 0.60	0.02 ± 0.02	0.70 ± 0.75	1.38 ± 0.82	2339.0 ± 938.9	228.85 ± 98.5
60 cm	0.06 ± 0.04	0.02 ± 0.02	1.41 ± 1.49	1.49 ± 1.50	1436.4 ± 870.0	69.10 ± 37.1
100 cm	0.76 ± 0.17	0.05 ± 0.01	3.30 ± 4.01	4.12 ± 3.63	1107.0 ± 483.8	6.42 ± 3.6

TOWeR, Timberlake Observatory for Wetland Restoration; PA, Preservation area; PP, Palmetto Peartree; DON, dissolved organic nitrogen; TDN, total dissolved nitrogen.

ments (Table 3). Drought treatment with SO_4^{2-} had lower nitrification potential than treatments without SO_4^{2-} (Fig. 5 and Table 3).

Discussion

Our results show that saltwater incursion into coastal wetlands leads to desorption and export of large quantities of reactive N, primarily as NH_4^+, and that the magnitude of this effect is magnified by past agricultural land use. We estimate that saltwater incursion facilitated by drought in 2007–2009 led to export of an additional 1579 kg NH_4^+, compared to years when salinity was lower (2009–2011, Table 2). We observed increases in NH_4^+ with salinization in all three coastal wetland sites (Fig. 3), but the magnitude was much lower in the established wetlands (PP and PA; highest concentrations ca. 1 mg l^{-1}) than in the restored wetland (TOWeR; up to 30 mg l^{-1} in soil solution). We suggest this disparity is largely due to the past agricultural land use of the TOWeR site as compared to PP and PA.

Most research to date on the biogeochemical consequences of saltwater incursion has focused on laboratory experiments (Weston *et al.*, 2006, 2011; Chambers *et al.*, 2011), different sites along salinity gradients (Craft, 2007; Krauss *et al.*, 2009; Giblin *et al.*, 2010), or artificial salt additions to field plots (Neubauer, 2013). This study provides the first evidence at the ecosystem scale of increased reactive N release due to drought-induced saltwater incursion. The linear increase in NH_4^+ concentrations with increasing Cl^- observed in our microcosm experiment indicates that much of the increased NH_4^+ release can likely be explained by dis-

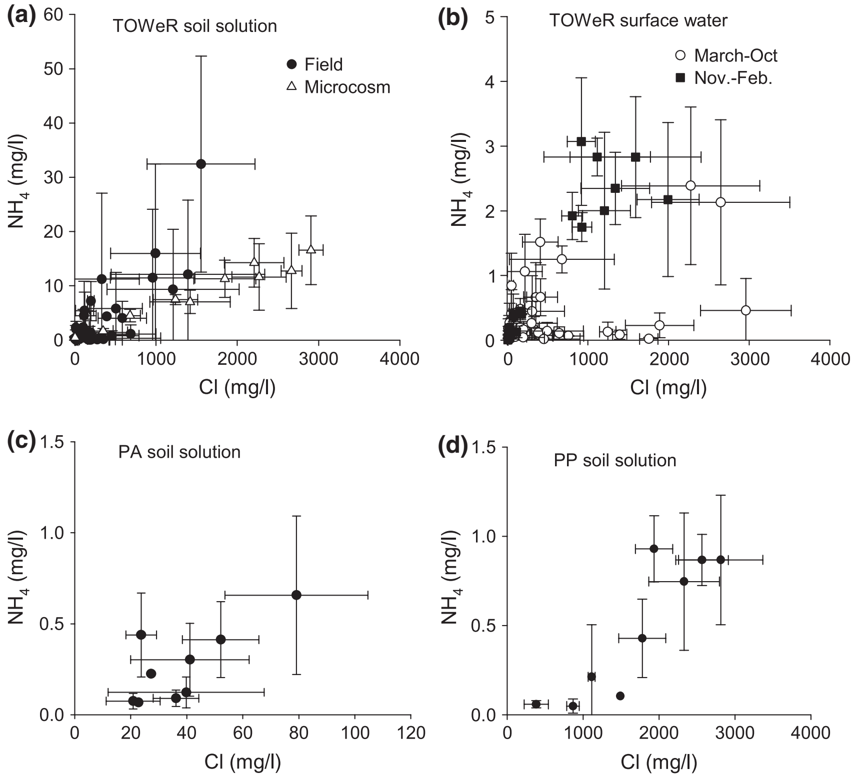

Fig. 3 Average Cl^- and NH_4^+ concentrations per sampling date in soil solution (a) and monthly concentrations in surface water (b) from the TOWeR site; and soil solution from Preservation area (c) and Palmetto Peartree Preserve (d). Panel (a) includes field soil solution data from TOWeR (black circles) and soil column experiment (white triangles). In panel (b) surface water data are separated into the growing season (March–October, white circles) and non growing season (November–February, black squares).

Table 2 Annual export and mean annual conductivity for 5 years for the Timberlake Restoration Project. DIN : DON ratio is the ratio of dissolved inorganic nitrogen (NO_3-N + NH_4-N) to dissolved organic nitrogen (DON)

Budget year	NH_4-N (kg ha^{-1} y^{-1})	NO_3-N (kg ha^{-1} y^{-1})	TDN (kg ha^{-1} y^{-1})	DON (kg ha^{-1} y^{-1})	DIN : DON ratio	Mean annual conductivity (ms cm^{-3})
2007–08	1.43	0.03	6.83	5.37	0.27	3.59
2008–09	4.35	0.12	6.78	2.77	1.61	4.92
2009–10	1.07	0.31	9.47	8.09	0.17	0.80
2010–11	1.12	0.20	9.05	7.73	0.17	0.49
2011–12	0.06	0.04	1.77	1.66	0.06	0.96

TDN, total dissolved nitrogen.

placement of NH_4^+ on cation exchange sites with saltwater base cations (Na^+, Ca^{2+}, Mg^{2+}, K^+, Fig. 3 and 4). Our results suggest that legacy fertilizer from past agricultural use exacerbates NH_4^+ release in response to salinization.

Our results suggest that increased N export from the surrounding landscape due to increased salinization could be very large. The TOWeR site is located in a coastal region that has had, and is predicted to have, high rates of sea-level rise compared to global averages (Sallenger *et al.*, 2012). Rising sea levels will increase salinity of the Albemarle Sound by facilitating the exchange between the estuary and the ocean (Corbett *et al.*, 2007). An estimated sea-level rise of up to 1 m in this region could lead to flooding of approximately 1661 km^2 of forested wetlands (Henman & Poulter, 2008) similar to our sites. There is approximately 149 Tg C in the soils of the Albemarle Peninsula that could be inundated by sea-level rise (Henman & Poulter, 2008). Assuming a C : N ratio of 21.4 and that an average 0.25% of the total N in the soils is exchangeable NH_4^+ (Morse & Bernhardt, 2013), we estimate

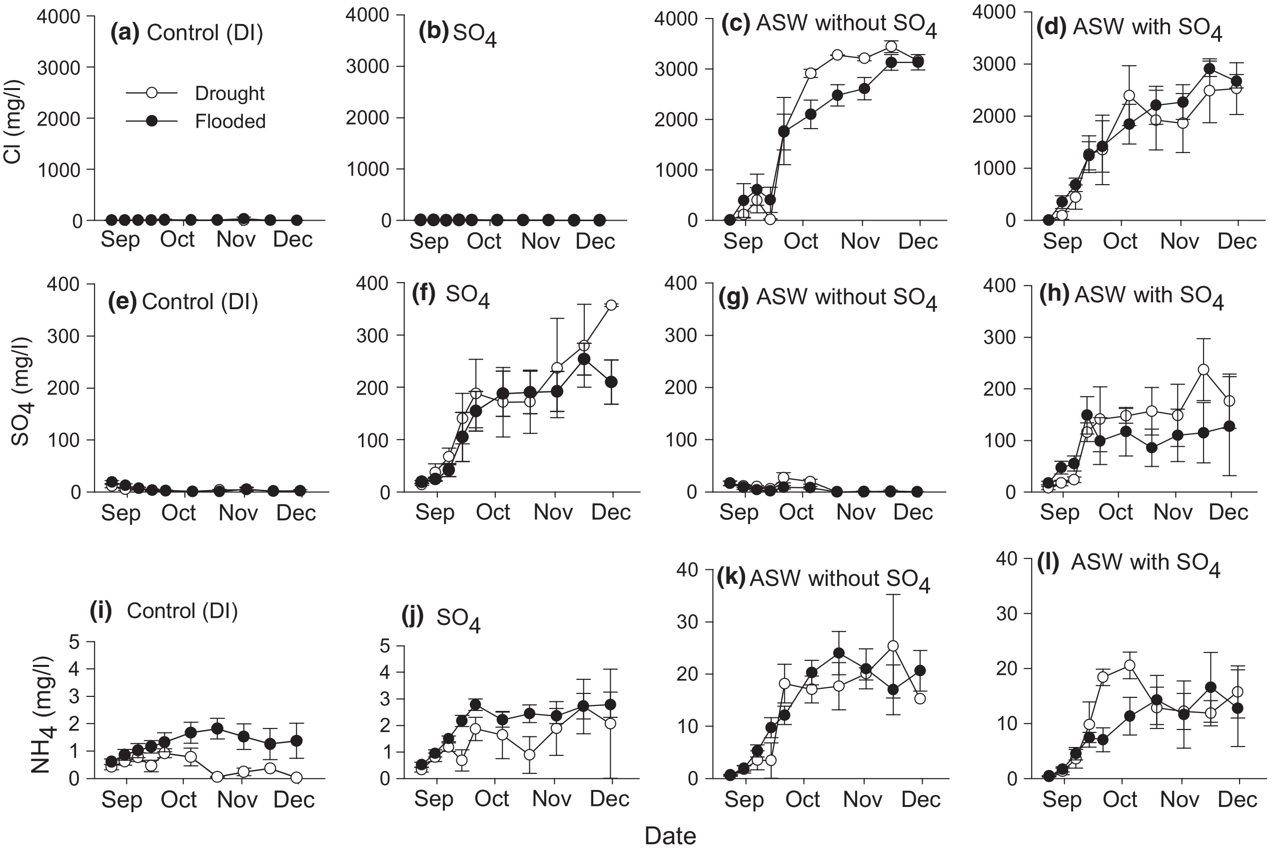

Fig. 4 Chloride (a–d), sulfate (e–h) and NH_4^+ (i–l) concentrations from soil solution (15 cm depth) from the soil column experiment. Hydrology treatments are differentiated by color (drought white circles, flooded black circles). Water chemistry treatments are arranged in increasing chemical complexity (control, SO_4^{2-} only, artificial seawater without SO_4^{2-}, and artificial saltwater with SO_4^{2-}). Note the different scales on *y*-axes on panels i–l.

Table 3 Results of ANOVA on log-transformed NH_4^+ from 15 cm piezometers and nitrification potential in microcosm experiment with hydrology (drought and flooded), salt (present or absent), and sulfate (present or absent) as factors

Parameter	df	Sum of squares	*F* ratio	*P*
NH_4^+				
Hydrology	1	75.87	27.40	**<0.0001**
Salt	1	289.56	104.57	**<0.0001**
SO_4	1	3.00	1.08	0.2986
Hydrology*Salt	1	18.09	6.53	**0.0111**
Hydrology*SO_4	1	2.28	0.82	0.3647
Salt*SO_4	1	26.34	9.51	**0.0022**
Hydrology*Salt*SO_4	1	0.73	0.26	0.6071
Nitrification potential				
Hydrology	1	1.22	41.9	**<0.0001**
Salt	1	0.05	1.73	0.19
SO_4	1	0.09	3.16	0.08
Hydrology*Salt	1	0.17	2.79	0.10
Hydrology*SO_4	1	0.08	6.00	**0.019**
Salt*SO_4	1	0.04	1.61	0.21
Hydrology*Salt*SO_4	1	0.01	0.49	0.48

Bold values indicate significant differences.

there are approximately 18 077 Mg NH_4^+ that could be released due to flooding associated with sea-level rise. To put that number in perspective, the Mississippi River (with a size almost 300 times the size of the watershed in which TOWeR site is located) delivers on average 31 000 Mg NH_4^+ per year to the Gulf Coast (Goolsby *et al.*, 1999). Our estimate of NH_4^+ that could be released due to flooding associated with sea-level rise represents an initial estimate based on our current knowledge of these systems. The time frame over which this stored N is likely to be released from saltwater-inundated soils will depend on the rate of sea-level rise, frequency of storms, severity of drought, and land-use decisions (particularly fertilizer application).

The timing of initial saltwater incursion into freshwater wetlands likely plays an important role in determining the consequences on N cycling. Because wetland restoration in this site required the cessation of active pumping and removal of water control structures, we were able to observe in this study the first instances of saltwater incursion in at least two decades at the TOWeR site. During those two decades, the site

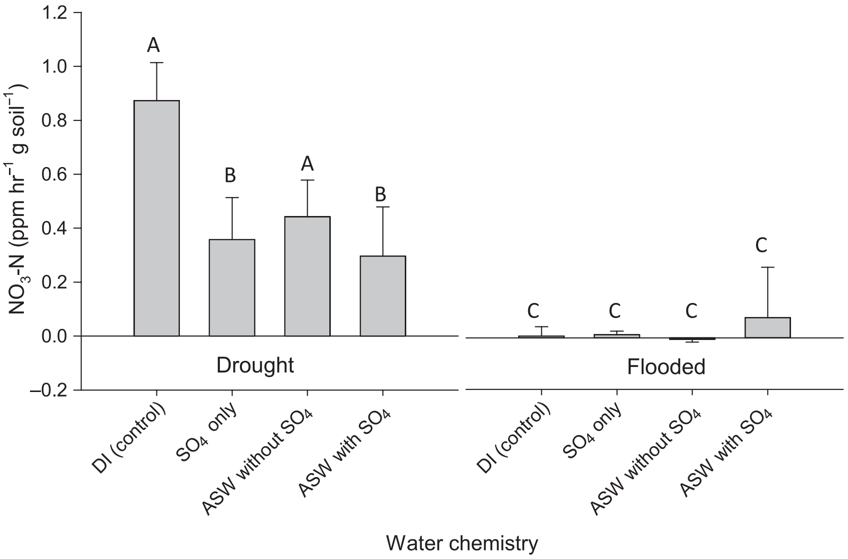

Fig. 5 Nitrification potential from soil column experiment measured at the end of the experiment. Different letters denote significant differences from ANOVA and post hoc Tukey on log-transformed data.

received significant inputs of fertilizer, leading to the accumulation of reactive N that could be released due to cation exchange with increased salinity. Our results differ from a recent study in tidal forests of Georgia and South Carolina, which did not find any increases in NH_4^+ release due to saltwater incursion (Noe *et al.*, 2013). Noe *et al.* (2013) hypothesized that because their study happened long after the first instances of saltwater incursion events, they had missed changes that might occur in the early events of saltwater incursion. However, our results from Palmetto Peartree site suggest that a site that continually receives low levels of salinity (commonly <5 ppt) could potentially release NH_4^+ for long periods of time. Noe *et al.* (2013) did find increased N mineralization rates under higher salinity that were attributed to increased inputs of labile detritus from dead and dying stressed trees, which might be occurring in the Palmetto Peartree site. We have observed increased frequency of tree mortality at our sampling site and other sections of the Palmetto Peartree site have been reported to have changed from forested to marsh (Poulter *et al.*, 2009).

In both surface water and soil solution from TOWeR, the increase in NH_4^+ concentrations due to salinity was nonlinear (Fig. 3a and b), this is in contrast to the linear increase observed in the microcosm experiment (Fig 3a). Nonlinear increases in field data has been also documented in a New England freshwater marsh that also experiences high salinity during low discharge (Giblin *et al.*, 2010). The lack of a simple linear relationship might be due to (i) time lags in the diffusion of saltwater into sediment and of NH_4^+ out of sediment; (ii) a nonlinear relationship between ion exchange of NH_4^+ and saltwater cations; and (iii) changes in available NH_4^+ due to the balance between generation (through mineralization) and removal or transformation of NH_4^+ (through plant uptake or nitrification). Lower NH_4^+ concentrations in surface waters may have been due to plant uptake and nitrification at the sediment-water interface (Fig. 3). Higher NH_4^+ concentrations in surface water with high salinity during the non growing season (November–February) than during the growing season (March–October), Fig. 3) and the time lag observed between increased Cl^- and NH_4^+ concentrations (Fig. S1) both suggest that plant uptake can mitigate salinity-induced NH_4^+ release.

Our experimental results suggest that increased salinity, particularly during a drought, could increase NH_4^+ release through both cation exchange and decreased nitrification. Coupled nitrification and denitrification is the primary mechanism of permanent N removal in this system (Morse & Bernhardt, 2013). Salinity has been shown to inhibit nitrification through increased toxicity of HS^- from increased SO_4^{2-} reduction (Joye & Hollibaugh, 1995) and osmotic stress on nitrifiers (Rysgaard *et al.*, 1999). In our soil column experiment, nitrification potentials were significantly higher in the drought cores than in the flooded treatments, in which nitrification potentials were near zero (Fig. 5). In the drought cores, the presence of SO_4^{2-} lowered nitrification potential compared to treatments without SO_4^{2-}, which could be due to toxicity of HS^- from increased SO_4^{2-} reduction (Joye & Hollibaugh, 1995; Fig. 5). Given that salinity can increase NH_4^+ release from sediments and decrease nitrification, these effects could synergistically increase NH_4^+ availability.

The combination of increased cation exchange and decreased nitrification can simultaneously exacerbate internal eutrophication problems for coastal wetlands and enhance N exports to estuaries.

In addition to the effects of salinity on nitrification, our results show that cation displacement by salts can have stronger effects on NH_4^+ fluxes in the short term than changes in microbial activity associated with increased SO_4^{2-} concentrations. Examining the concentrations of Cl^- and SO_4^{2-} as they diffuse across this coastal landscape suggests that the spatial extent of increased salt cations (Cl^-, Na^+, Mg^{2+}, Ca^{2+}) might be greater than the spatial extent of elevated SO_4^{2-} concentrations. Cl^- ions and their attendant base cations move conservatively upstream, while SO_4^{2-} is preferentially consumed by sulfate-reducing microbes. The Cl : SO_4 ratio thus increases with distance from the saltwater source (Fig. S3). Thus, we would expect a larger area of coastal wetlands to be impacted by saltwater-mediated ion exchange than by the biological effects of increased SO_4^{2-}, though much of the research on the consequences of saltwater incursion to date has focused on the effects of increased SO_4^{2-}.

Release of NH_4^+ through cation exchange due to increased salinity might be more predictable and easier to model than changes mediated by vegetation and microbial activity (Weston *et al.*, 2010). Saltwater-induced changes in N cycling due to altered microbial metabolism will be more difficult to predict, because they will be determined by a combination of negative effects of ionic stress, stimulatory effects of increased availability of SO_4^{2-}, inhibitory effects of HS^-, reductions in dissolved carbon availability, and the pace of microbial community adaptation. Understanding the relative importance of abiotic cation exchange and biotically driven changes will be increasingly important in managing coastal water quality, as saltwater incursion becomes more widespread with enhanced drought severity and sea-level rise.

Our results raise important questions regarding the long-term efficacy of large-scale wetland restoration programs seeking to reduce N loading to estuaries. Plans to implement wetland restoration projects throughout coastal zones should consider whether hydrologic reconnection is likely to increase the frequency and intensity of saltwater incursion and the potential for such changes to constrain wetland N retention. While hydrologic reconnection is an important factor in the recovery of ecosystem function lost to past wetland conversion, it is important to both consider and plan for negative consequences associated with changing environmental conditions, such as our observed saltwater-induced NH_4^+ release.

Coastal land development, dredging, human water use, drought, and accelerated sea-level rise will all make saltwater incursion into low-lying coastal areas more common and more severe. Our results suggest that these same land cover and climate alterations are likely to contribute to coastal wetland, marsh, and estuarine eutrophication, even in the absence of additional reactive N loading to these systems. Recent work has shown that increased N loading can accelerate loss of marsh ecosystems (Deegan *et al.*, 2012), highlighting the importance of understanding salinity-induced N release from coastal wetlands to downstream salt marshes. Previous research on the consequences of saltwater incursion on carbon cycling in coastal wetlands has focused on large changes in plant communities and productivity, such as the regime shift from forested wetlands to marshes. Our results show large changes to the N cycle of coastal wetlands in the early stages of saltwater incursion, prior to major changes in plant community composition. Understanding changes in N cycling of these coastal wetlands is critical to prevent further N pollution to coastal ecosystems.

Acknowledgements

We thank M. Burke-Scoll, K. Balance, and A. Fedders for help in the field and in the laboratory. K. Krauss and two anonymous reviewers provided helpful comments on earlier versions of this manuscript. This research was supported by GDSMB private gift in support of basic research, a grant to E.S. Bernhardt from the US Department of Energy's Office of Science (BER) through the Coastal Center of the National Institute for Climatic Change Research at Tulane University, and NSF DEB-1021149. M. Ardón was supported by NSF DBI-0805576 and DBI-1216512. J.L. Morse was supported by a U.S. Environmental Protection Agency Science to Achieve Results (STAR) Fellowship (FP16599).

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Diagram of microcosm setup explaining location of samplers and reservoir used to manipulate water chemistry and water table elevation.
Figure S2. Weekly NH_4^+ concentrations (mg l^{-1}, dark circles), Cl^- (mg l^{-1} white circles, first right axis) and salinity (ppt, white circles, second right axis) at the Outflow of TOWeR for 5 years.
Figure S3. Weekly Cl^- vs. SO_4^{2-} concentrations (mg l^{-1}) from three sites across TOWeR. All regressions were significant at $P < 0.05$.