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RESEARCH ARTICLE

Fertilizer legacies meet saltwater incursion: challenges and constraints for coastal plain wetland restoration

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Coastal wetland restoration is an important tool for climate change adaptation and excess nutrient runoff mitigation. However, the capacity of restored coastal wetlands to provide multiple ecosystem services is limited by stressors, such as excess nutrients from upstream agricultural fields, high nutrient legacies on-site, and rising salinities downstream. The effects of these stressors are exacerbated by an accelerating hydrologic cycle, expected to cause longer droughts punctuated by more severe storms. We used seven years of surface water and six years of soil solution water chemistry from a large (440 ha) restored wetland to examine how fertilizer legacy, changes in hydrology, and drought-induced salinization affect dissolved nutrient and carbon concentrations. To better understand the recovery trajectory of the restored wetland, we also sampled an active agricultural field and two mature forested wetlands. Our results show that nitrogen (N) and phosphorus (P) concentrations in soil solution were 2–10 times higher in the restored wetland compared to two mature forested wetlands, presumably due to legacy fertilizer mobilized by reflooding. Despite elevated nutrient concentrations relative to reference wetlands, the restored wetland consistently attenuated N and P pulses delivered from an upstream farm. Even with continued loading, N and P concentrations in surface water throughout the restored wetland have decreased since the initial flooding. Our results suggest that high nutrient concentrations and export from wetlands restored on agricultural lands may be a severe but temporary problem. If field to wetland conversion is to become a more widespread method for ameliorating nutrient runoff and adapting coastal plain ecosystems to climate change, we should adopt new methods for minimizing the initial export phase of wetland restoration efforts.

Keywords: nitrogen; phosphorus; sea-level rise; wetlands; eutrophication; drought

Introduction

Wetland restoration is becoming an important tool to counteract coastal degradation and enhance the provision of ecosystem services (Silliman et al., 2015). Restored wetlands can ameliorate nutrient runoff to sensitive coastal ecosystems (Zedler, 2003), and wetland restoration efforts are increasingly being used to mitigate or adapt to sea-level rise and increased frequency of severe storms (Jones et al., 2012; Temmerman et al., 2013). Economic incentive programs for wetland restoration have been suggested as cost- effective ways to deal with excess nutrients and recover other ecosystem services provided by wetlands (Zedler and Kercher, 2005). However, most restored wetlands have reduced diversity and provide fewer or less efficient ecosystem services than their natural wetland counterparts, even decades after restoration (Craft et al., 2003, Ballantine and Schneider 2009; Moreno-Mateos et al., 2012). Incomplete recovery of function in restored wetlands can lead to unforeseen negative environmental consequences. For example, when wetland restoration is conducted in former agricultural fields, fertilizer legacies can limit the recovery of phosphorus (P) retention and lead to elevated P export (Ardón et al., 2010a; Ardón et al., 2010b; Kinsman-Costello et al., 2014).

Agricultural expansion has led to the loss of many wetlands globally (Verhoeven et al., 2006), and efforts to reverse this trend restore wetland hydrology to agricultural areas. Wetlands reestablished in former fields or pastures may be influenced by the legacies of agricultural use for decades or centuries after the practices have ceased (Foster et al., 2003; Potter et al., 2004). Agricultural legacies alter carbon and nutrient pools in soils (McLauchlan, 2006), vegetation succession of old fields (Cramer et al., 2008), and the restoration trajectory of lakes (Bennett et al., 1999). But it is not clear how agricultural legacies alter the

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recovery of restored wetlands. Agricultural disturbances such as draining, soil compaction, decreased soil permeability, loss of soil organic matter, changes in local topography, and fertilizer application can impair recovery of wetland functions (Bruland et al., 2003; Steven and Gramling, 2012). The time period over which these legacies affect ecosystem function after the cessation of practices differs depending on the type of disturbance (Bain et al., 2012). For example, a meta-analyses of wetland restoration suggested that the recovery of hydrologic functions likely occurs within the first five years after restoration, while full recovery of biological structure and biogeochemical processes can remain incomplete even a century after restoration (Moreno-Mateos et al., 2012). It is important to understand how long agricultural legacies can affect the function of restored wetlands, given that they can reduce the capacity of these ecosystems to respond to rising sea levels and a changing climate.

Wetland restoration efforts are further complicated by the need to consider shifts in climate that may constrain the potential for projects to achieve their ecological goals. While there is disagreement in the exact magnitude of changes in weather at regional scales, many areas will experience changes in extreme events (IPCC 2014). The southeast US is likely to experience increased frequency and duration of droughts punctuated by more severe storms (Carter et al., 2014). Increased frequency of storms will mobilize accumulated nutrients in sediments (Kaushal et al., 2014). In contrast, increased frequency of droughts in the southeast coastal plain will facilitate the landward movement of saltwater into former freshwater areas (Ardón et al., 2013). This increase in salinity could lead to increased release of both nitrogen and phosphorus from soils and sediments. This occurs primarily in two ways: 1) high concentrations of divalent cations (Ca^{2+} and Mg^{2+}) in saltwater displace NH_4^+ from soil cation exchange sites (Weston et al., 2010; Ardón et al., 2013); and 2) the high concentration of sulfate in saltwater stimulates sulfate-reducing bacteria in anoxic sediments, resulting in the production of sulfide. Sulfide, in addition to being a potent phytotoxin (Lamers et al., 2013), suppresses nitrification (Joye and Hollibaugh, 1995), denitrification (Seitzinger et al., 1991) and outcompetes P (PO_4^{3-}) ions binding with iron (Caraco et al., 1989; Lamers et al., 1998). Increased sulfate from salinity also increases microbial carbon mineralization (Weston et al., 2011), which could lead to P release from organic sources (Chambers et al., 2011). It is unclear how increased frequency and duration of flooding with elevated salinity could affect the timing and magnitude of legacy N and P release from restored wetlands (Herbert et al., 2015).

The combination of agricultural legacies, a changing climate, and rising sea-levels are creating novel biogeochemical regimes in coastal plain wetlands (Helton et al., 2014). In addition to these stressors, restored wetlands can receive high nutrient loading from upstream agricultural fields (**Figure 1**). Our understanding of the recovery trajectory of restored wetlands under these novel biogeochemical regimes is limited due to the lack of long-term studies. We previously reported that drought-induced saltwater incursion led to increased inorganic N release from natural and restored wetlands (Ardón et al., 2013), and decreased export of dissolved organic carbon (Ardón et al., 2016). Here, we examined the long-term patterns in nutrients and carbon concentrations in surface water and soil solution of a restored wetland seven years after the initial reflooding. We also sampled an active agricultural field and a mature forested wetland to better understand the recovery trajectory. We asked: 1) how have surface water and soil solution concentrations of N and P changed seven years after reflooding the restored wetland? 2) How does drought-induced salinity alter the concentrations of N and P in an active agricultural field, a mature forested wetland, and a restored wetland? And 3) What are the main environmental drivers of N and P concentrations in an active agricultural field, a mature forested wetland, and a restored wetland? We hypothesized that N and P concentrations in soil solution and surface water would decline in the restored wetland as legacy nutrients are exported, but that this gradual decline in concentrations would be punctuated by drought-induced salinity pulses of nutrient export.

Methods

Site description

We focused on a large scale restored wetland in the coastal plain of North Carolina, USA. The Timberlake Observatory for Wetland Restoration (TOWeR) is located in Tyrell County

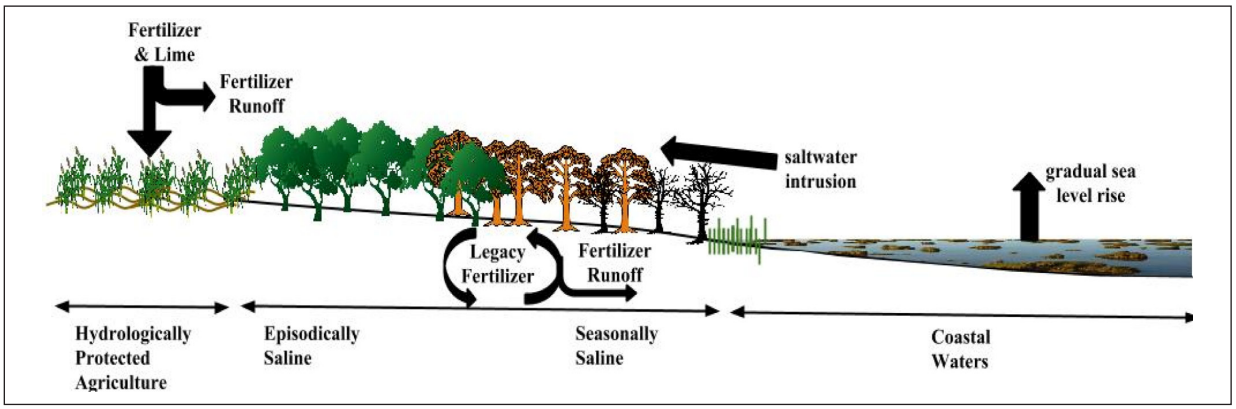

Figure 1: Conceptual diagram of upstream and downstream stressors affecting wetland restoration sites in the coastal plain of the US. DOI: https://doi.org/10.1525/elementa.236.f1

(35° 54′22″N, 76° 09′25″W, **Figure 2**). The restoration area is part of the Great Dismal Swamp Mitigation Bank, LLC, a compensatory mitigation bank that includes: 420 ha of mature forested wetland, 787 ha of forested wetlands, 57.2 of drained shrub-scrub, and 440 ha of former agricultural fields undergoing stream and wetland restoration. The two major soil series at the site are Ponzer muck (loamy, mixed, dysic, themirc Terric Haplosaprists) and Hyde loam (fine-silty, mixed active, thermic Typic Umbraquults, USDA SSURGO database 2005). Like much of the surrounding landscape, this area is flat with elevations ranging from −1 to 2 m above sea level (Lidar survey by the National Center for Airborne Laser Mapping 2008, Houston, Texas, USA). A detailed description of the history of the site and restoration can be found in Ardón et al. (2010a). The site drains to the Little Alligator River, and then the Albemarle Sound. The Albemarle Sound is part of the second largest estuarine system in the US. Due to the limited exchange with the ocean, it does not experience lunar tides, but does experience wind-driven tides. Salinity is in the brackish range [0–7 parts per thousand (ppt) (Corbett et al., 2007)], but during drought years salinities as high as 12 ppt have been measured in the Albemarle Sound (Division of Marine Fisheries, North Carolina Department of the Environment and Natural Resources, personal communication).

Soil solution sampling

To examine potential N and P release from the sediment to surface water, we measured soil solution every 2–6 months on 15–38 sampling points across the elevation gradient at the restored wetland (TOWeR) and two mature forested

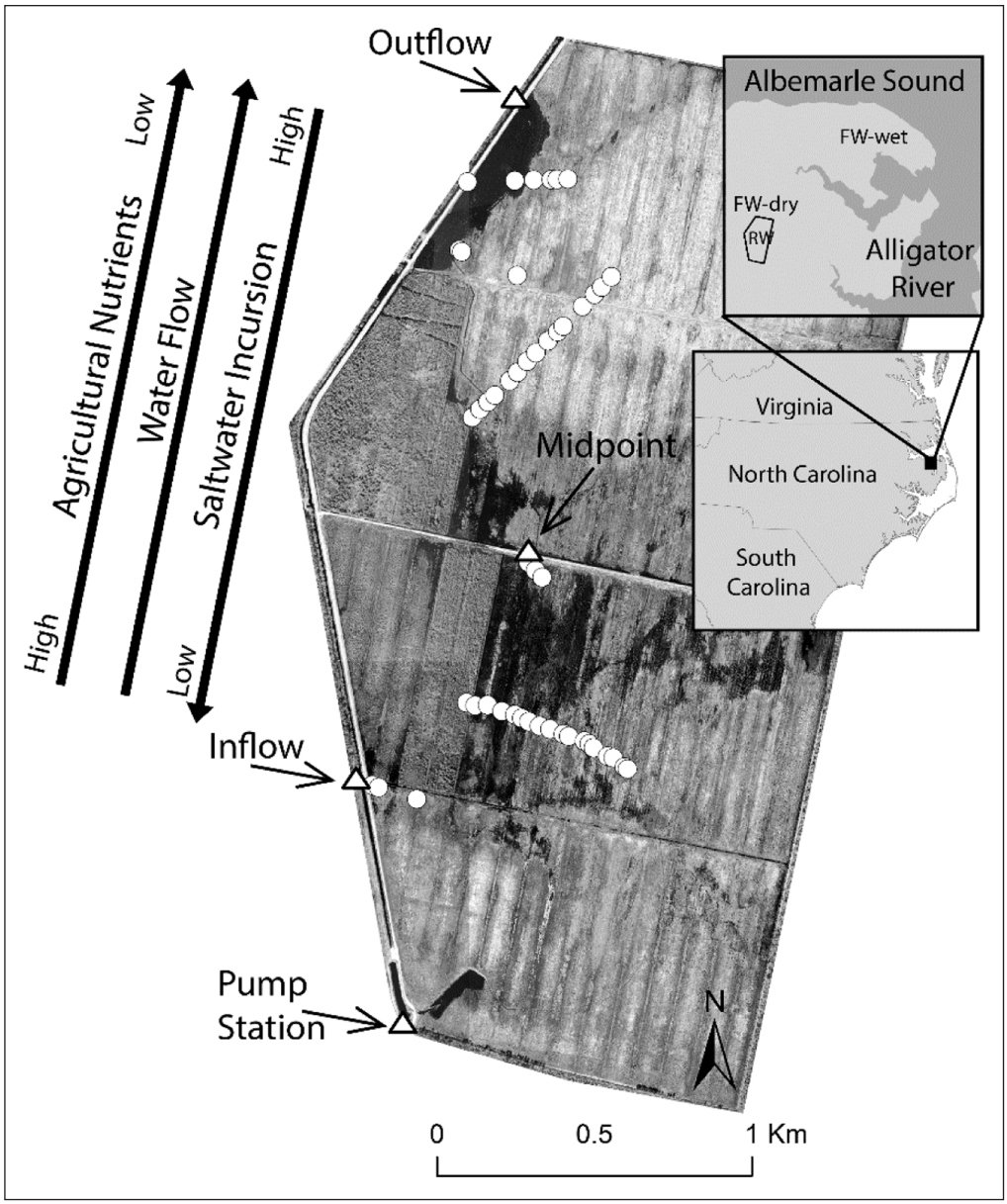

Figure 2: Map of the TOWeR site. Insert shows location in North Carolina and in the Albemarle-Pamlico Peninsula. Large black arrows indicate predominant direction of water flow, direction of saltwater incursion, and gradient in nutrients from agricultural runoff. Circles indicate soil solution and triangles indicate surface water sampling sites. DOI: https://doi.org/10.1525/elementa.236.f2

wetlands. One of the mature wetlands is within the TOWeR complex: FW-dry. We also sampled soil solution at Palmetto Peartree Preserve, which is located 8 km away from TOWeR. It is adjacent to the Albemarle Sound and low elevation, making it wetter than the other mature forested wetland, so we refer to it as FW-Wet (in agreement with sites described in Morse et al., 2012). Because of its location, FW-wet also frequently experiences higher salinities than TOWeR. Within TOWeR, we sampled 33 sampling points every 2–3 months for the first three years. During the last three years of the study we focused on a subset (15 sites) that still covered the hydrologic and salinity gradients across the site (**Figure 2**) and sampled every 3–6 months. Samples were collected from piezometers (wet conditions) and lysimeters (drier conditions) at 15 cm depth. For the two mature forested wetlands we sampled 5 sites in each one during the first three years of the study. Samples were collected by purging the piezometers, or adding a vacuum (40 kPa) on the lysimeters, and then collecting the samples after 24 hours. Samples were filtered in the field (Whatman GF/F, 0.7 μm) and collected in HDPE bottles and frozen until analyses. Soluble reactive phosphorus (SRP) was measured using the ascorbic acid and molybdenum blue method (APHA 1998). NH_4–N was measured using the phenate method on a Lachat QuickChem (Lachat Instruments, Milwaukee WI) automated system. NO_3–N, Cl^- and SO_4^{2-} were measured on a Dionex ICS-2000 ion chromatograph (Dionex Corporation, Sunnyvale, California, USA). Dissolved organic carbon (DOC) and total dissolved nitrogen (TDN) were measured on a Shimadzu TOC-V total carbon analyzer with a TNM–1 nitrogen module (Shimadzu Scientific Instruments, Columbia, Maryland, USA). Dissolved organic nitrogen was estimated as the difference between TDN and the sum of inorganic N (NO_3–N and NH_4–N).

Surface water monitoring

We sampled water elevation from four sites within the restored wetland: Pump Station, Inflow, Midpoint, and Outflow (**Figure 2**). The Pump Station drains a 2424 ha active farm undergoing corn and soybean rotation. The Inflow drains 420 ha of mature forested wetland. The Midpoint and the Outflow drain the upper and lower areas of the restored wetland (**Figure 2**). We recorded water depth and velocity every 15 minutes from 25 February 2007 to 27 February 2014 using acoustic Doppler area velocity meters (2150 Area velocity Meter, Teledyne ISCO, Lincoln, Nebraska, USA, **Figure 2**). We sampled culverts draining each of these land uses in order to be able to use the known area of the culvert, water depth, and velocity to estimate discharge. The active agricultural field is drained by two pumps on opposite ends, we sampled the pump that drains into the restored wetland (**Figure 2**). The pump was only activated after large rain events. We also measured rainfall using tipping bucket rain gauges (0.01 inch rain tip gauge, Teledyne ISCO, Lincoln, NE, USA).

Weekly surface water samples from all four sites were collected during the same time period as water flow measurements. Samples were filtered immediately (GF/F Whatman filters, 0.7 μm) and frozen until analyses. Solutes were analyzed as described above (*Soil solution sampling*).

Statistical analyses

We used ANOVA and post-hoc Tukey comparison on log transformed data to compare concentrations of surface water and soil solution among sites. To examine long-term increases or decreases in surface water concentrations we used non-parametric Kendall tau tests because they are better suited for the nature of time series data (Kendall, 1938). We also used Principal component analyses (PCA) to examine changes in water chemistry across sites and over time. To visualize changes in water chemical variability we used chull() to outline the convex hull of PCA results. A convex hull is the smallest convex polygon in a plane that contains all of the points in a given dataset.

We used mutual information (MI) to assess the degree to which nutrient concentrations covary with environmental factors (Scheuerell, 2017). Based upon the entropy of two or more random variables, MI is a nonparametric way of characterizing the mutual dependence between them (Cazelles, 2004; Gouhier and Guichard, 2014). Specifically, given two marginal entropies $H(X)$ and $H(Y)$, and their joint entropy $H(X,Y)$, then the normalized (0–1) mutual information between X and Y according to (Forbes, 1995) is:

$$MI(X,Y) = \frac{H(X) + H(Y) - H(X,Y)}{\sqrt{H(X)H(Y)}}$$

An MI value of 0 means that knowing everything about one time series would tell you absolutely nothing about the second, just a when Pearson's rho is zero. Conversely when MI equals 1, the temporal dynamics of both time series are identical. Using MI is particularly appealing in a case like this where the data may be highly skewed or zero-inflated because unlike parametric correlation coefficients (e.g., Pearson's), MI makes no assumptions about the distributional form of the variables. We determined the MI among solutes (nutrients and DOC) and drivers (water level, chloride, and precipitation) within each site at time lags of –4 to 4 weeks. We explored the different time lags to examine the possibility that solutes could be behaving similarly (similar increases and decreases in concentration), but at different time lags. A negative lag means X (chloride, level, precipitation) leads Y (solutes), a positive lag means X lags behind Y. We previously reported that NH_4^+ concentrations often lagged behind Cl^- concentrations by a month (Ardón et al., 2013). We used Monte Carlo resampling of the original time series (100) to determine whether the calculated MI was significant at each lag (see Cazelles, 2004). All statistical analyses were conducted in version 3.3.2 of the R software (R Team 2016) or JMP (SAS, Cary, NC).

Results

Soil solution

Chloride and sulfate concentrations were highest in one of the forested wetlands (FW-Wet, Cl^- range 300–1800 mg/L, SO_4^{2-} range 100–200 mg/L) than in the other sites (Cl^- range 20–1200 mg/L, $F_{ratio} = 68.14$, $p < 0.001$, SO_4^{2-} range 5–150 mg/L, $F_{ratio} = 46.63$, $p < 0.001$, **Figure 3A** and **B**). Chloride and sulfate concentrations in TOWeR were highest in 2008, which was the year with most severe saltwater

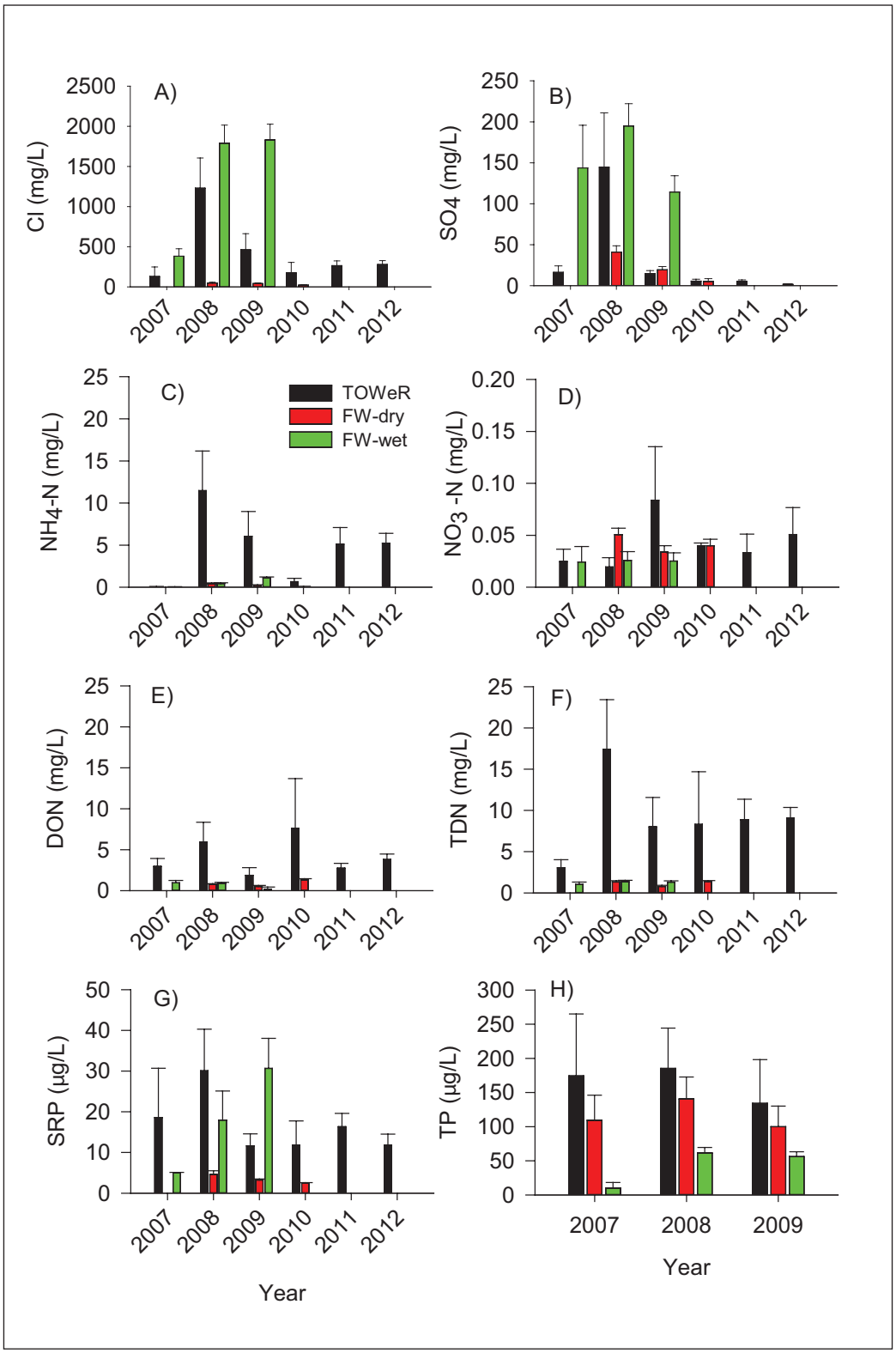

Figure 3: Average (+ standard error) concentrations of chloride **(A)**, sulfate **(B)**, ammonium **(C)**, nitrate **(D)**, dissolved organic N **(E)**, total dissolved N **(F)**, soluble reactive phosphorus **(G)**, and total phosphorus **(H)** in soil solution in one restored wetland (TOWeR) and two mature forested wetlands (FW-Dry and FW-Wet). DOI: https://doi.org/10.1525/elementa.236.f3

incursion (**Figure 3A** and **B**). Ammonium concentrations were up to ten-fold higher in TOWeR (mean in 2011 was 11.4 mg/L NH_4–N, range 0.06 to 11.4 mg/L) compared to the two forested wetlands (range 0.06–1 mg/L, $F_{ratio} = 11.17, p < 0.0001$, **Figure 3C**). NO_3–N concentrations were low across all three sites (range 0.005–0.08 mg/L, **Figure 3D**). Total dissolved N and dissolved organic N (DON) were both significantly higher in the restored wetland than in the two mature forested wetlands (TOWeR TDN mean = 9.6 mg/L compared to 1–1.3 mg/L in the two forested sites, $F_{ratio} = 18.75$, $p < 0.001$; TOWeR DON mean = 3.5 mg/L compared to 0.5–0.60 mg/L for the two

forested sites, F_{ratio} = 14.07, p < 0.001; **Figure 3E** and **F**). SRP concentrations were higher in FW-Wet (mean SRP = 23 ± 3.2 μg/L) and TOWeR (mean SRP = 16.1 ± 2.1 μg/L) compared to FW-Dry (mean SRP 3.7 ± 3.8 μg/L, F_{ratio} = 7.52, p < 0.001, **Figure 3G**). TP was higher in the restored wetland (mean TP = 149.3 ± 24.6 μg/L) and FW-dry (mean TP = 130.1 ± 29.9 μg/L) than FW-wet (mean TP = 58 ± 25.4, F_{ratio} = 3.56, p < 0.05, **Figure 3H**). The year with the highest SRP in soil solution at TOWeR was 2008, which was also the year with the highest Cl^- concentrations (**Figure 3A** and **G**), since then SRP has declined (**Figure 3G**).

Surface water elevation and precipitation

Water elevation at the Pump Station was measured at the end of a large pump which is elevated, thus the high (1.5 m) baseline (**Figure 4**). The deviations from that

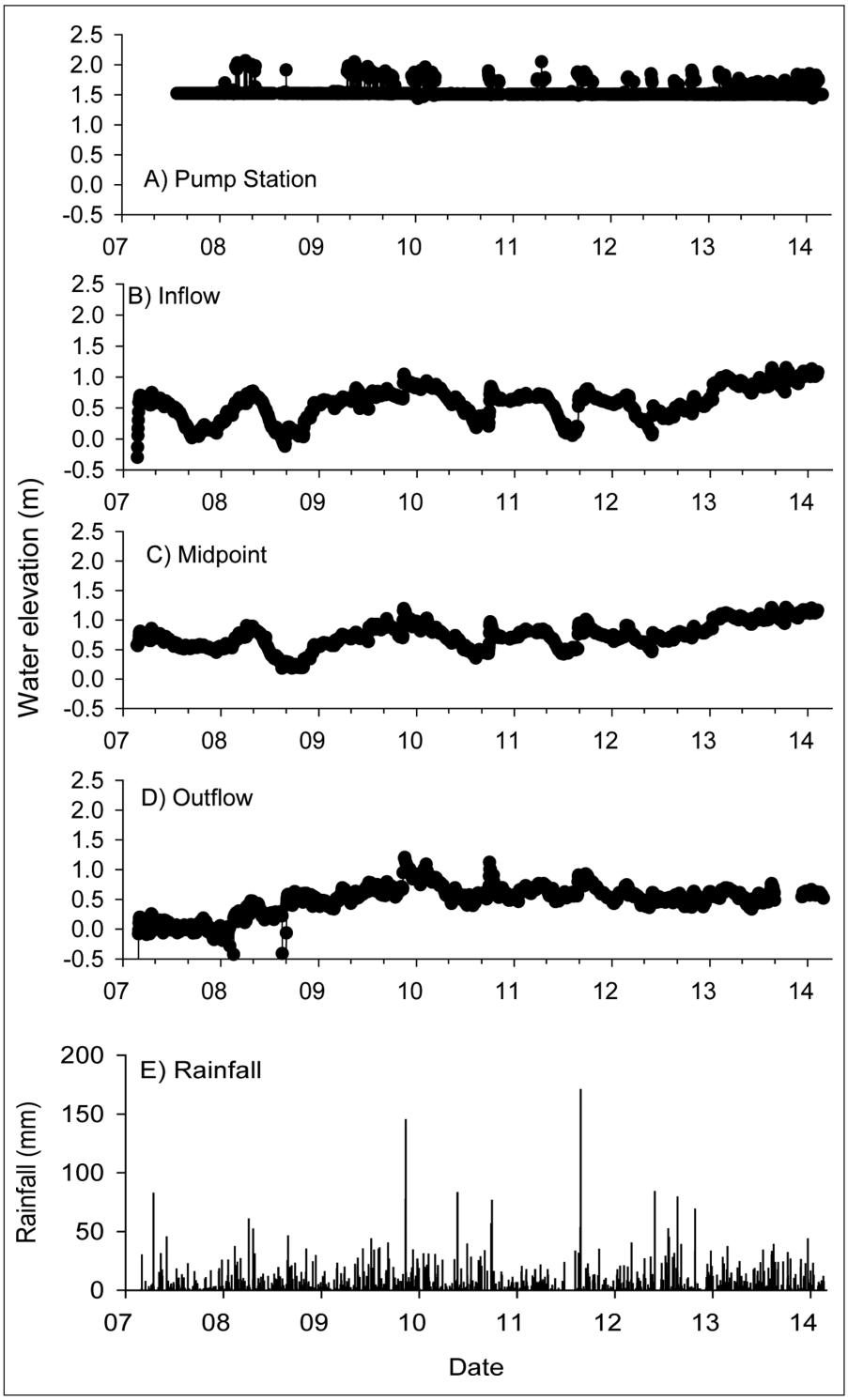

Figure 4: Daily average water surface elevation (m above NGVD29) for the four surface water sampling sites **(A–D)** and precipitation for the site **(E)**. The sensor for the Pump Station site is located within a large elevated pipe that is only turned after rain events. DOI: https://doi.org/10.1525/elementa.236.f4

1.5 m are when the pump was activated. Water elevation at the Inflow, Midpoint, and Outflow has increased over time, while it decreased in the Pump Station (**Table 1**, **Figure 4**). Water level in the Inflow, Midpoint, and Outflow all decreased during periods of low precipitation (**Figure 4**). The Outflow water level shows two drawdown experiments in 2008, in which we turned on the pump that used to drain the entire site and decreased water level to approximately 10 ha of the site (Ardón et al., 2010b). There was no long-term trend in precipitation over the study period.

Surface water

Surface water chemistry varied in space and time. Saltwater incursion during drought periods led to Cl^- concentrations being highest in the Outflow, and declined moving upstream, with the lowest concentration in the Pump Station (**Figure 5**, **Table 2**). Chloride concentrations were highest during the drought periods in 2007 and 2008, and declined over the study period in all four sites (**Table 1**). Sulfate concentrations were significantly higher in the Outflow and the Pump Station, compared to the other two sites (**Table 2**). Sulfate concentrations declined in all sites over time (**Table 1**), and behaved similarly to Cl^- concentrations in all sites, except the Pump Station where SO_4^{2-} was higher than we would have expected based on the Cl^- concentrations (**Figure 5**).

Inorganic phosphorus concentrations (SRP) were highest in the Outflow (**Table 2**), and declined in all four sites over the study period (**Figure 6**, **Table 1**). Repeated pulses of SRP at the Pump Station were attenuated as water move through the site, for example look at P pulse in fall 2010 at Pump Station, but small increases at the Outflow (**Figure 6**). SRP at the Outflow has been around the detection limit since the summer of 2012. NH_4–N concentrations were highest in the Outflow and lowest in the Pump Station (**Table 2**). NH_4–N concentrations declined in all sites except the Pump Station, where there were no significant changes over time (**Figure 7**, **Table 1**). NH_4–N concentrations were highest during the 2007 and 2008 drought-induced saltwater incursion events (**Figures 5** and **7**). NO_3–N concentrations were highest in the Pump Station and exhibited seasonal increases of up to 6 mg/L NO_3–N. However, those spikes in nitrate where attenuated through the site and concentrations were not significantly different among the other sites (**Figure 7**, **Table 2**). There were no significant increase or decrease in NO_3–N concentrations in any of the sites over the study period (**Table 1**). TDN concentrations were highest in the Inflow and lowest in the Midpoint (**Table 2**). TDN concentrations declined in the Inflow, Midpoint, and Outflow, but showed no change in the Pump Station (**Figure 8** and **Table 1**). DON concentrations were highest in the Inflow and lowest in the Pump Station (**Figure 8** and **Table 2**). There were no significant changes in DON concentrations over time in any of the sites. DOC concentrations were highest in Inflow and lowest in the Pump Station (**Table 2**). DOC concentrations increased over time in the Inflow and Outflow (**Figure 9**).

Mutual information analyses showed that there was strong synchrony between chloride and SRP, DOC, DON, TDN, and NH_4–N in the Inflow, Midpoint, and Outflow (**Figure 10**), suggesting that salinity is an important environmental driver of nutrients at various time lags (–4 to 4 weeks). Water level also had high mutual information with SRP, DOC, TDN, NH_4–N, and NO_3–N in the Inflow, Midpoint, and Outflow, though the MI values were lower than for Cl^- (**Figure 10**). At the Pump Station, water level was the environmental driver that most commonly exhibited high MI with DOC, DON, NH_4–N, and NO_3–N (**Figure 10**). The significant MI values at multiple time lags suggests high autocorrelation within each of the time series, which is evident in the time series plots (**Figures 5**–**9**). Precipitation patterns showed low MI with nutrient and DOC concentrations.

Principal component analyses of the water chemistry for all sites and all seven years explained 33.9% of the variation in principal component 1 and 23.2% in principal component 2. PCA 1 was positively correlated with Cl^- (r = 0.49), SO_4^{2-} (0.47), and NH_4–N (0.38) and negatively correlated with DOC (–0.44). PCA 2 was positively correlated with TDN (0.63), DON (0.48), and NH_4–N

Table 1: Kendall Tau tests for water level and monthly solute concentrations during 2007–2014 for each of the four sites in TOWeR.[a] DOI: https://doi.org/10.1525/elementa.236.t1

	Pump Station	Inflow	Midpoint	Outflow
Water level	–0.177	0.311	0.385	0.295
SRP	–0.3	–0.144	–0.187	–0.311
NH_4–N		–0.267	–0.335	–0.357
NO_3–N				
TDN		–0.211	–0.403	–0.293
DOC		0.251		0.161
DON				
Cl^-	–0.163	–0.359	–0.465	–0.399
SO_4	–0.358	–0.446	–0.46	–0.472

[a]Negative values indicate long-term decline, while positive values indicate long-term increase. All are values present are significant at $p < 0.05$.

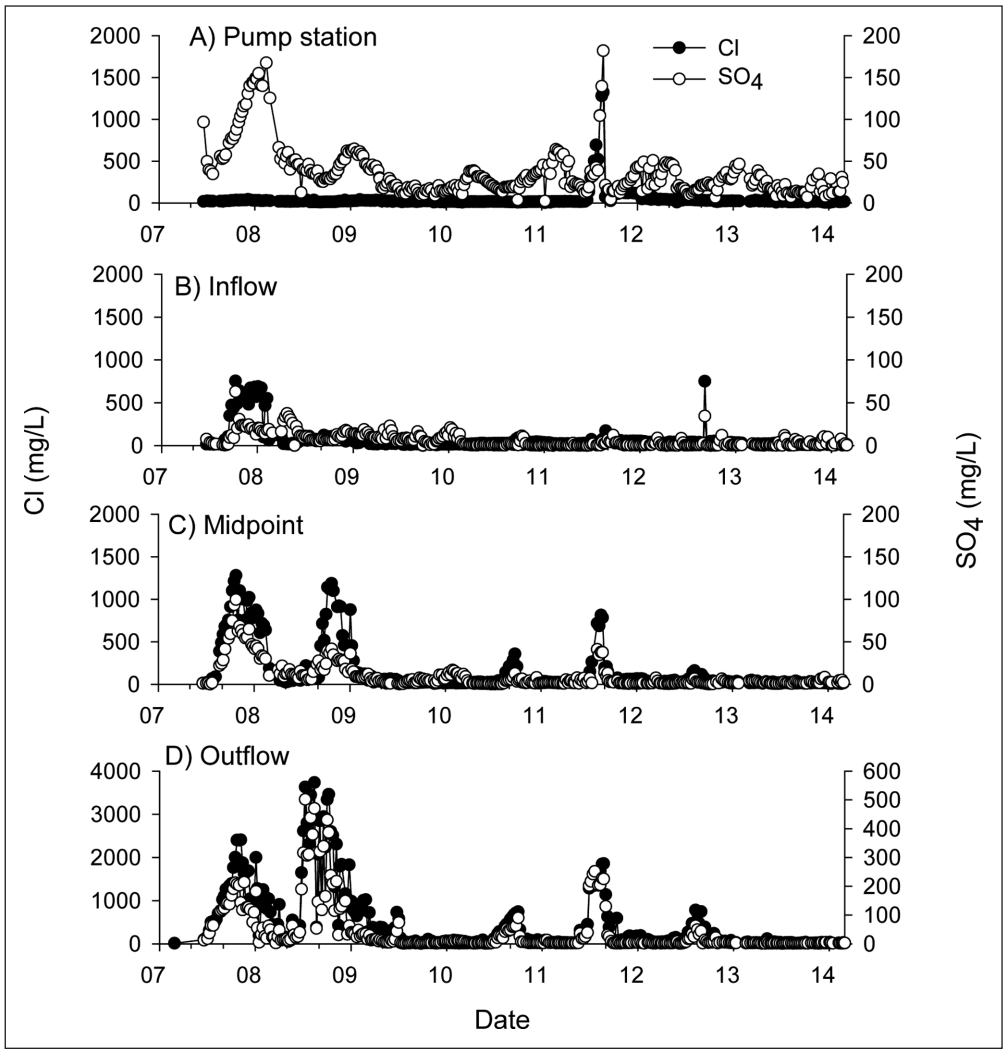

Figure 5: Seven years of weekly surface water concentrations of chloride (black circles) and sulfate (white circles) in four sites in TOWeR. Note the different scale on y-axes in panel D for both Cl^- and SO_4). DOI: https://doi.org/10.1525/elementa.236.f5

Table 2: Average (standard error) solute concentrations in surface water at four sites in TOWeR. DOI: https://doi.org/10.1525/elementa.236.t2

Site	SRP	NH_4–N	NO_3–N	TDN	DON	DOC	Cl^-	SO_4
Pump Station	0.006 c	0.25 c	0.51 a	1.63 b	0.92 c	26.37 c	43.89 d	34.61 a
	(0.001)	(0.02)	(0.05)	(0.05)	(0.02)	(0.55)	(6.60)	(1.55)
Inflow	0.007 b	0.38 b	0.11 b	1.84 a	1.35 a	43.59 a	67.57 c	5.72 b
	(0.001)	(0.04)	(0.02)	(0.05)	(0.03)	(0.88)	(7.21)	(0.42)
Midpoint	0.006 bc	0.36 b	0.04 b	1.59 b	1.21 b	35.36 b	146.82 b	8.88 b
	(0.001)	(0.05)	(0.01)	(0.05)	(0.02)	(0.73)	(14.83)	(0.81)
Outflow	0.008 a	0.49 a	0.04 b	1.70 ab	1.18 b	33.25 b	434.34 a	40.26 a
	(0.001)	(0.05)	(0.01)	(0.05)	(0.03)	(0.70)	(38.25)	(4.35)

[a] All concentrations in mg/L. Letters denote significant differences ($p < 0.05$) based on ANOVA and post-hoc Tukey on log transformed data.

(0.43). Variability in water chemical parameters has changed little in Pump Station during the 7 years of the study, as shown by similar range in PCA values in Y1 and Y7 (**Figure 11A**). In contrast, the Inflow, Midpoint and Outflow had much higher variability in Y1 than in Y7 of the study (**Figure 11B, C, D**), driven primarily by the

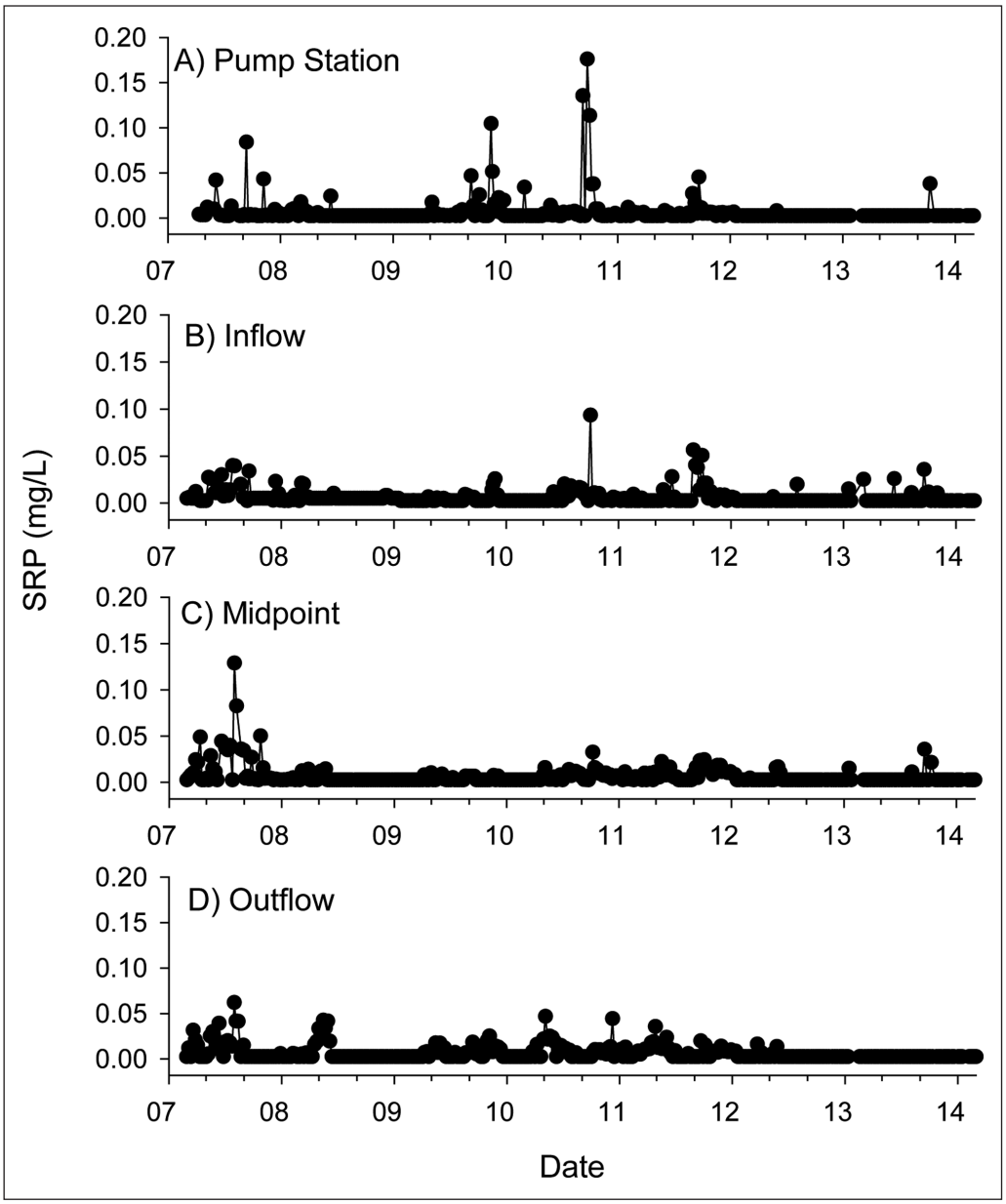

Figure 6: Seven years of weekly surface water concentrations of soluble reactive phosphorus (SRP) in four sites in TOWeR. DOI: https://doi.org/10.1525/elementa.236.f6

water chemistry changes associated with saltwater incursion during the first year of the study.

Discussion

As we anticipated, we found that nutrient concentrations in the restored wetland were consistently higher than in two nearby forested wetlands, but that the magnitude of these differences have declined over time. Despite fertilizer legacies, the restored wetland has attenuated pulses on N and P from an adjacent farm. Surface water concentrations of N and P have declined faster than soil solution, suggesting the importance of monitoring both surface water and soil solution. We conclude that the observed long-term declines in N and P result from the rather rapid export of fertilizer legacies, suggesting that elevated nutrient export from restored wetlands is likely to be a temporary problem.

We previously hypothesized that this site could release P for 5–15 years after reflooding, based on the first two years of data collection (Ardón et al., 2010b). The longer period in this dataset shows that SRP concentrations in surface water at the Outflow have mostly returned to regional background concentrations six years after the initial reflooding (**Figure 6**). NH_4–N concentrations in surface water have also declined since the beginning of our study (**Table 1**), but this decline is mostly driven by the large pulses in NH_4–N concentrations caused by drought-induced saltwater incursion during the first 2 years of the study (Ardón et al., 2013). Soil solution NH_4–N concentrations have not declined as fast as surface water (**Figures 3** and **7**). Drought-induced saltwater incursion led to increased concentrations of both surface water and soil solution NH_4–N. The high mutual information between chloride and N in all sites (**Figure 10**) within the restored

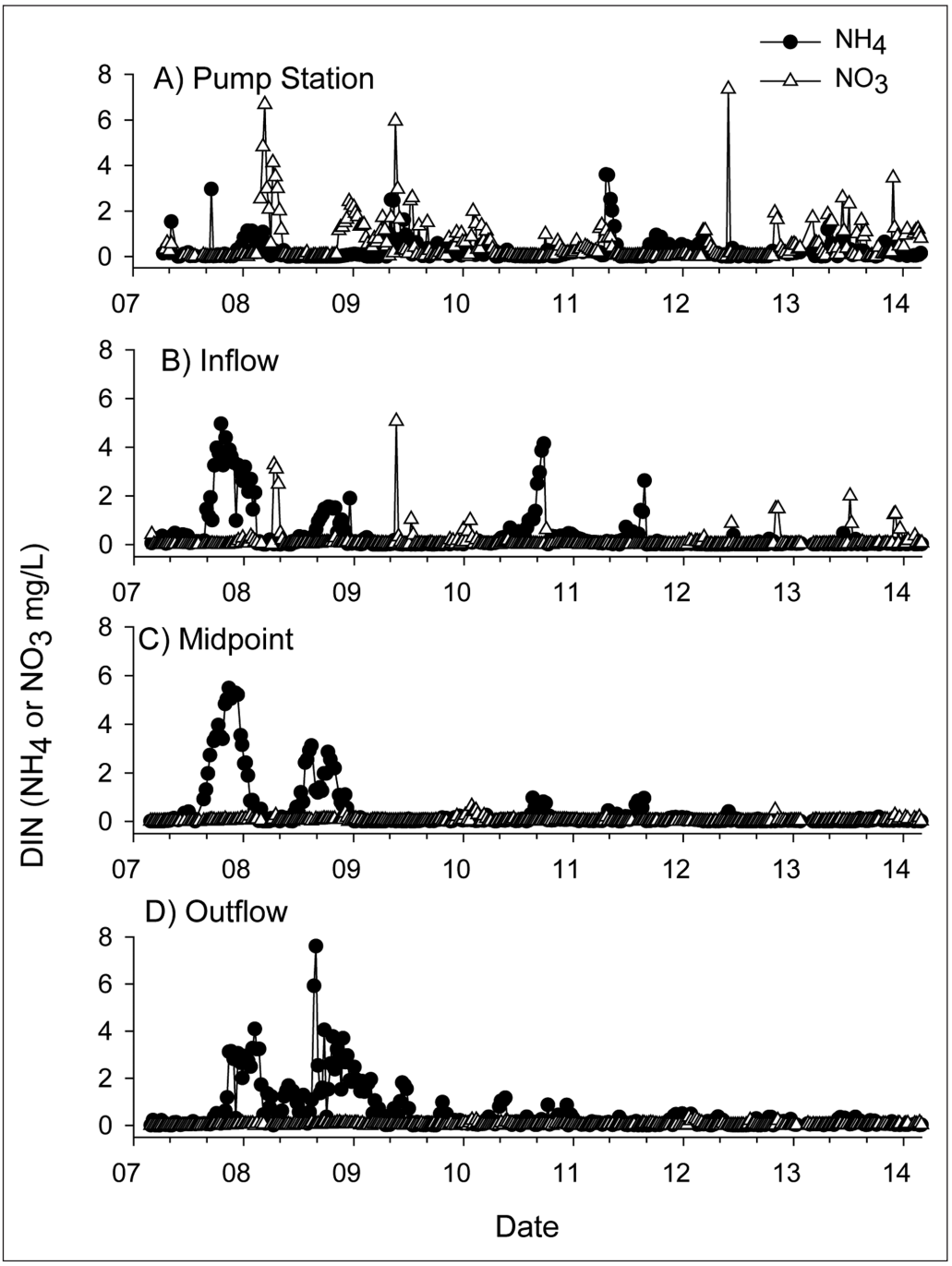

Figure 7: Seven years of weekly surface water concentrations of ammonium (black circles) and nitrate (white circles) in four sites in TOWeR. DOI: https://doi.org/10.1525/elementa.236.f7

wetland indicate that salinity is an important environmental driver of N concentrations. Given that the site has not experienced other saltwater incursion events as large as in the first two years of our record, we cannot forecast if NH_4–N concentrations will increase as much if the site experienced another event of similar magnitude, but the high NH_4–N concentrations in soil solution suggest there is a potential for NH_4–N release. Our results suggest that nutrient legacies in former agricultural fields should be incorporated into planning monitoring timetables for wetland restoration projects, given that it could be a problem for a minimum of six years after initial restoration.

Novel biogeochemical regimes

The combination of agricultural legacies, current agricultural practices, and saltwater incursion events can lead to novel biogeochemical regimes that are highly chemically variable in space and time (**Figures 1** and **11**). Similar to novel ecosystems, which consist of combinations of species that have no analogue in nature (Hobbs et al., 2006), the relative concentrations and their temporal and spatial variability in these novel biogeochemical regimes like do not have analogues in natural ecosystems. It is possible that these novel combinations of solutes will alter the structure and function of restored wetlands.

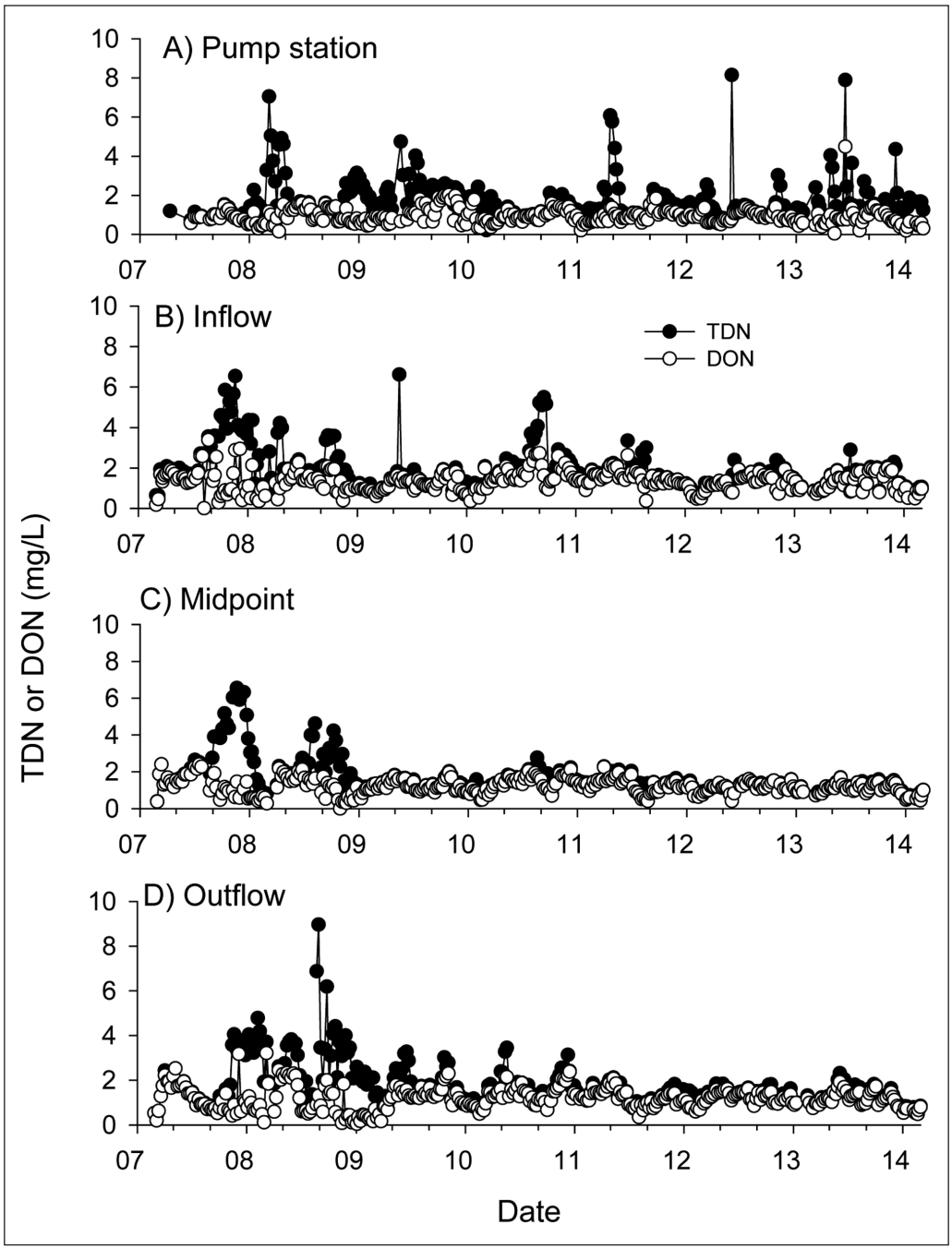

Figure 8: Seven years of weekly surface water concentrations of total dissolved N (black circles) and dissolved organic N (white circles) in four sites in TOWeR. DOI: https://doi.org/10.1525/elementa.236.f8

For example, the high chloride concentrations during saltwater incursion events increased ammonium and decreased DOC concentrations (Ardón et al., 2013, 2016), and decreased bald cypress growth (Powell et al., 2016). These changes in the availability of carbon (C) and N can alter the stoichiometric balance needed for microbial growth and the availability of electron donors and acceptors for microbes (Helton et al., 2015). Changes in the C to N ratio of dissolved organic matter (DOM) observed could affect the bioavailability of DOM to microbial communities at the site and downstream estuary (Ardón et al., 2016). The decline in the variability in water chemistry in the restored wetland between years 1 and 7 evidenced in the PCA analyses (**Figure 11B, C,** and **D**), suggests that these highly heterogeneous biogeochemical regimes might fade as ecosystems recover through restoration. We need a better understanding of how changes in the relative availability of C and nutrients, and electron donors and acceptors, will alter biogeochemical cycling in wetlands experiencing agricultural activities and rising salinities.

Rising salinities are not the only source of ions that can alter biogeochemical pathways. Our results show that chloride and sulfate concentrations can become decoupled due to human activities. The Pump Station site had higher sulfate than would be expected based on the chloride concentrations (**Figure 5**). We believe the high sulfate

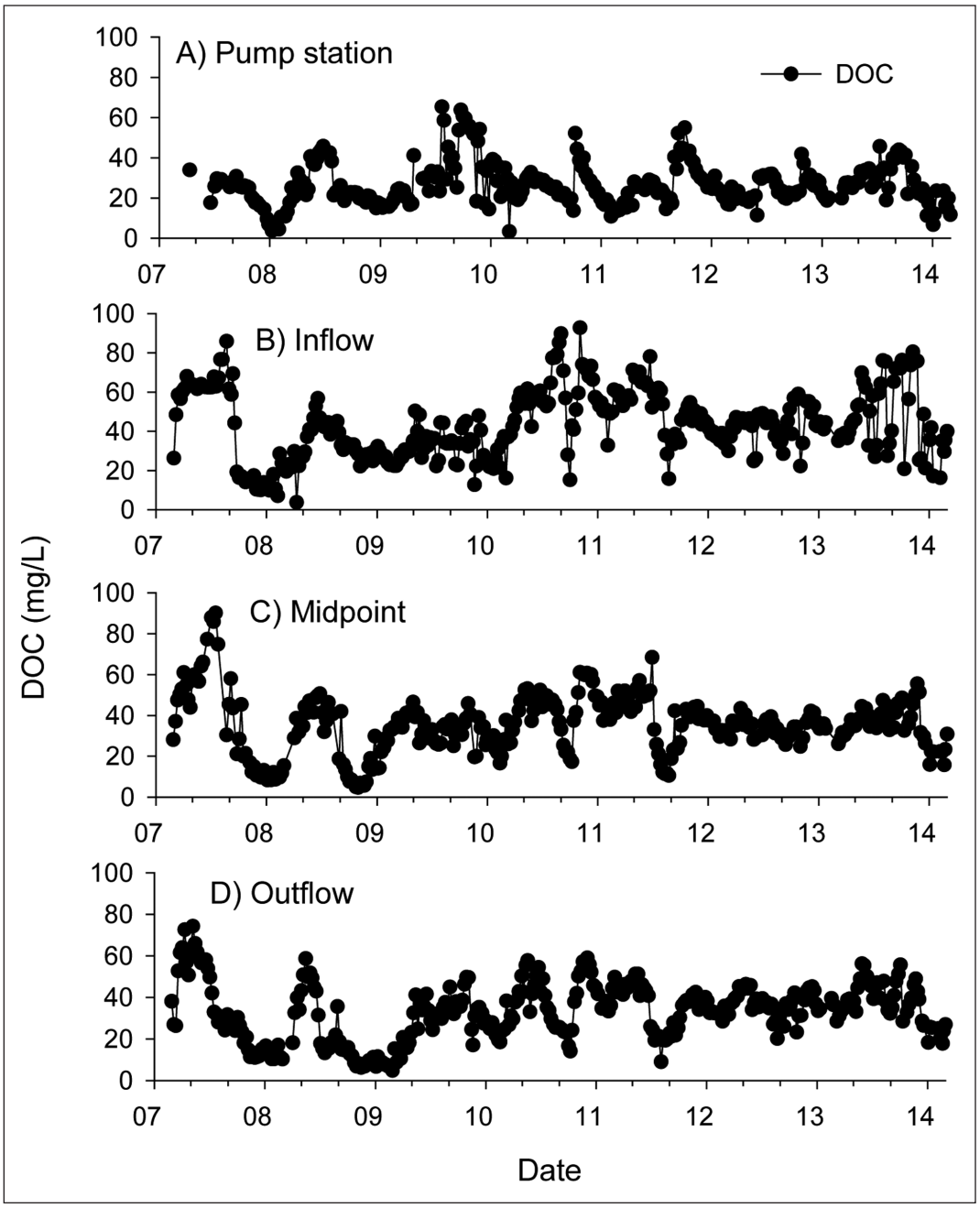

Figure 9: Seven years of weekly surface water concentrations of dissolved organic C (DOC) in four sites in TOWeR. DOI: https://doi.org/10.1525/elementa.236.f9

comes from agricultural runoff (potentially gypsum or pesticides) from the farm adjacent to our site (Helton et al., 2014). Increased sulfate concentrations can increase sulfate reduction and decrease methanogenesis (Helton et al., 2014). These results illustrate the importance of understanding the relative concentrations of different ions within saltwater and run-off from human activities, because ions can exhibit different behavior and have different biogeochemical consequences (Cañedo-Argüelles et al., 2016).

The combination of rising salinities and agricultural legacies we documented in this site are likely to be present in other restored wetlands. About half of the wetland restoration projects in the southeastern US occurred in areas with an agricultural past (Steven and Gramling, 2012). Our review of the ongoing wetland restoration projects in the coastal counties of North Carolina suggest that up to 70% of the area that has undergone wetland restoration used to be in agricultural production (DENR personal communication). Thus, the same patterns we have observed in the TOWeR site are likely occurring throughout the southeastern US. Studies have shown that agricultural P legacies will limit wetland restoration in the Everglades for the next 50–120 years (Reddy et al., 2011). There are also plans for large scale wetland restoration in Louisiana and Maryland, places with large expanses of agricultural lands. At this point it is unclear how much of this wetland restoration will occur in former agricultural fields, but given the historical importance of agriculture to the economy of all of these states, many of the wetland

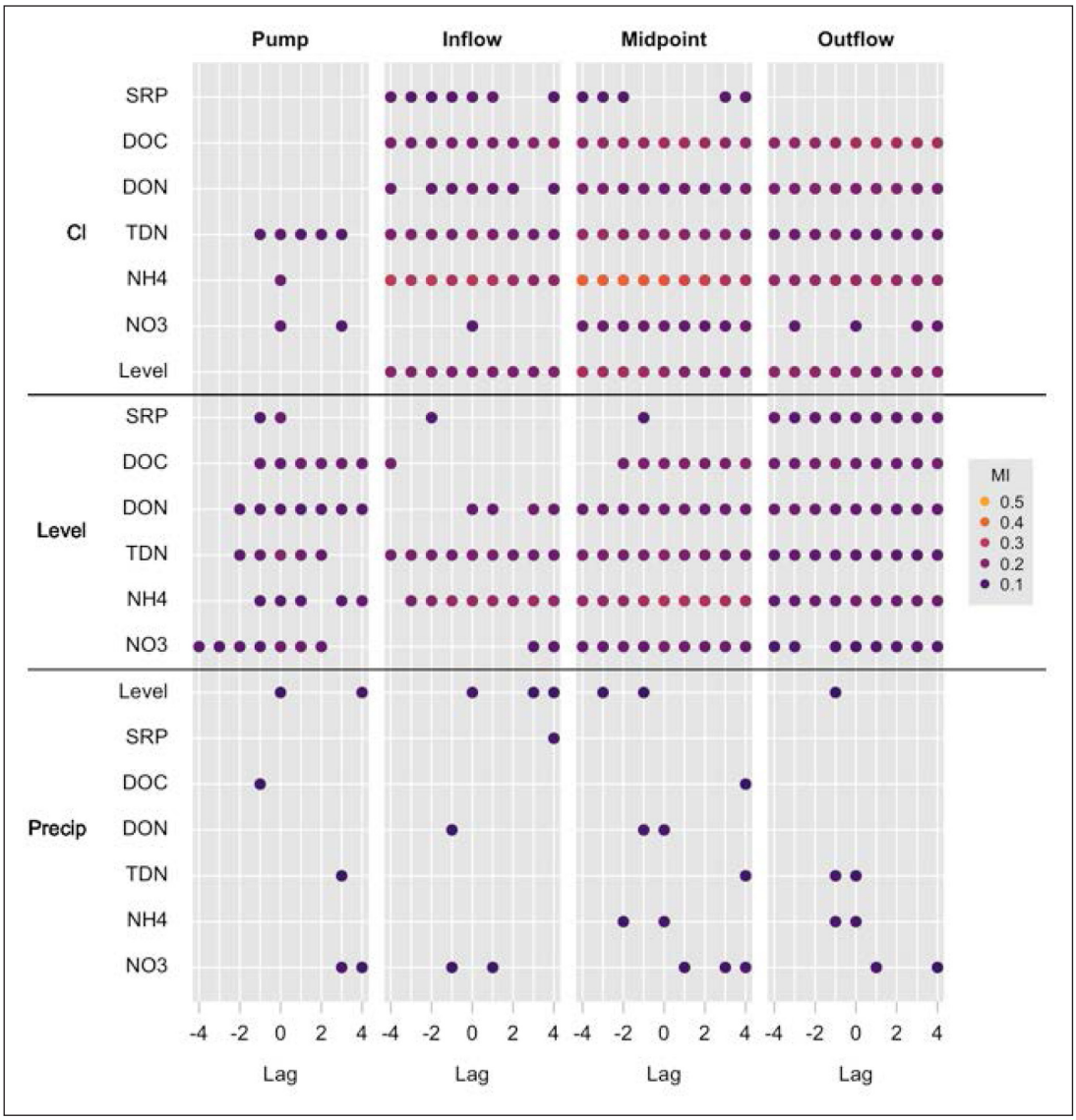

Figure 10: Mutual information results at various time lags from (–4 to 4 weeks) for environmental drivers (chloride, water level, and precipitation) and water solutes. Points indicate significant MI, the color refers to MI value scaled to 1.0, but the highest values we observed were 0.5. DOI: https://doi.org/10.1525/elementa.236.f10

restoration projects will likely occur in former agricultural fields. Wetland restoration in former agricultural fields in coastal areas is also occurring in Canada (Bortolotti et al., 2016), Spain (Calvo-Cubero et al., 2014), and the Netherlands (Van Dijk et al., 2004). Our results suggest that agricultural legacies in restored wetlands that are vulnerable to increased salinization could have a significant impact on downstream estuaries in all these areas, but differences in duration of agricultural practices, soil types, and hydrology will affect the magnitude and timing of nutrient release.

While the potential for legacy P to continue to cause eutrophication problems even after decreased loading has been well recognized in the literature (Jarvie et al., 2013; Sharpley et al., 2013), only recently has it been documented at large scales for N (Van Meter et al., 2016). Legacy N could potentially remain in soils for up to 35 years after stopping agricultural practices in the Mississippi basin (Van Meter et al., 2016). While we observed significant declines in surface water N, soil solution concentrations have not declined as fast, suggesting that there is still the potential for legacy fertilizer to be exported. Our previous work has also shown that legacy N in the restored wetland leads to elevated N release during saltwater incursion events (Ardón et al., 2013). Some of that legacy N will likely be transformed to N_2 gas due to increased denitrification from reflooding. In our site, coupled nitrification and denitrification are the most important forms of N loss to the atmosphere (Morse and Bernhardt, 2013), suggesting that places in the landscape with alternating oxic/anoxic conditions might be hot spots of N transformation. We need more long-term studies that follow the fate of N in wetland restoration sites to better constrain the timing and magnitude of legacy N release.

In contrast to N, P in surface water did not appear to respond to increased salinity. Our results show that drought-induced saltwater incursion caused increases in soil solution P, but not in surface water. The highest P

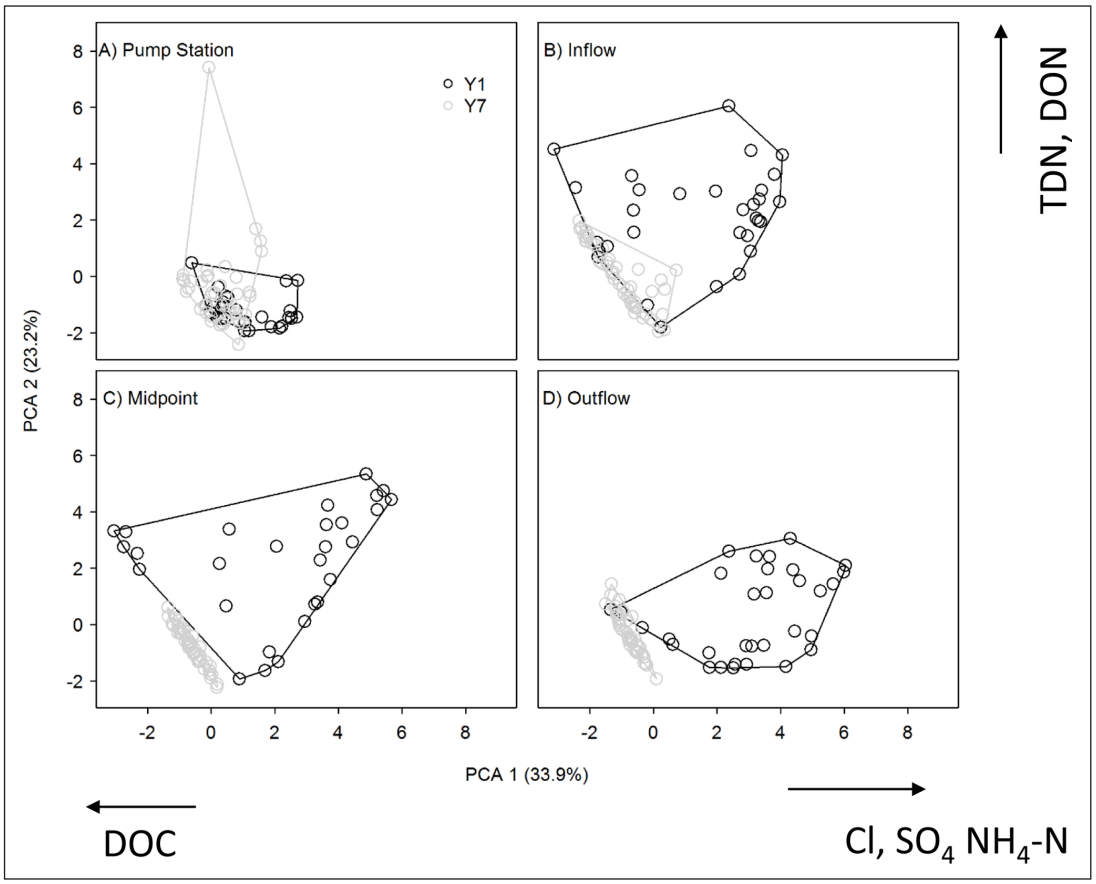

Figure 11: Principal component analyses of surface water chemistry for years 1 and 7 for the Pump Station **(A)**, Inflow **(B)**, Midpoint **(C)**, and Outflow **(D)**. PCA 1 explained 33.9% and PCA 2 explained 23.2% of the variation. Lines are convex hulls connecting the outer points of each dataset. Dark colors are for year 1 and light colors are for year 7 for each site. DOI: https://doi.org/10.1525/elementa.236.f11

concentrations in soil solution in TOWeR were in 2008, which was the year with highest saltwater incursion (**Figure 3A** and **G**). However, we did not observe elevated P concentrations in surface water during saltwater incursion events. Previous studies have shown increased P release in soil solution due to increased salinity (Sundareshwar and Morris, 1999; Chambers et al., 2011; Van Diggelen et al., 2014). The prevailing paradigm has been that increased salinity leads to increased availability of P due to sulfide binding to Fe, leading to the release of P, and increasing mineralization of organically bound P. Increases in sediment P availability has been seen with increasing salinity in Chesapeake Bay (Hartzell and Jordan, 2012). However, other studies have reported decreased P availability with increasing salinity (Baldwin et al., 2006; van Dijk et al., 2015). Two plausible reasons we did not observe increased P in surface water are that increased plant uptake prevented the extra soil solution P from leaving the site in surface water, or that salt-induced flocculation of dissolved organic carbon immobilized P (Ardón et al., 2016). Previous work has shown that salt induced flocculation can lead to the immobilization of P (Sholkovitz, 1976). Our results suggest that P export from the site after the initial reflooding could have been higher, had the site not experienced severe drought and saltwater incursion. Increasing frequency and duration of high salinity flooding of former freshwater wetlands will make it challenging to predict the net effect of wetland restoration on water quality.

Implications for wetland restoration

Wetland restoration will play an increasingly important role in increasing the resilience of our coast (Sutton-Grier et al., 2015). Our results suggest that we need a more sophisticated understanding of the landscape context when deciding where and when to place wetland restoration projects. First, it is important to understand both the upstream and downstream potential stressors. Using wetland restoration to ameliorate nutrient runoff in agricultural watersheds means that these restored wetlands will receive high concentrations of nutrient runoff. There has been much research showing that high nutrient loading leads to decreased plant and animal diversity (Verhoeven et al., 2006), and that continued loading may saturate the capacity of restored wetlands to retain nutrients, a critical ecosystem service (Richardson, 1985). It is also important to understand the downstream sources of stressors to these coastal wetland ecosystems. In our restored wetland, the removal of water control structures for hydrologic reconnection with the downstream Albemarle Sound was done to facilitate the recolonization of river herring (*Alosa aestivalis* and *Alosa pseudoharengus*). However, this also

facilitated the landward movement of marine salts during extended droughts (Ardón et al., 2013). With the goal of facilitating migration of diadramous species, hydrologic reconnection has also allowed saltwater to enter the site during droughts. For wetland restoration to be an efficient tool in the mitigation of excess nutrient runoff and climate change adaptation, we should adopt new methods for minimizing the initial nutrient release phase of wetland restoration efforts.

Data Accessibility Statement

The data presented here will be made available through Dryad Digital Repository.

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Competing interests

The authors have no competing interests to declare.

Author contributions

- Contributed to conception and design: MA and ESB
- Contributed to acquisition of data: MA and AHM
- Contributed to analyses and interpretation of data: MA, AHM, MDS, ESB
- Contributed to writing and revising: MA, AHM, MDS, and ESB

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