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Terrestrial freshwater influences salt marsh nitrogen processing through plant productivity

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Abstract

Tidal marshes serve as control points for nitrogen (N) removal at the terrestrial-marine interface. However, marshes are inherently heterogeneous, making it challenging to predict the timing, magnitude, and distribution of N processing. We investigated the role of terrestrial freshwater inputs in driving N-cycling in a tidal marsh dominated by a single plant species (*Salicornia pacifica*) (Elkhorn Slough National Estuarine Research Reserve). At this site, elevation-driven differences in tidal inundation and freshwater inputs have influenced the timing and distribution of *S. pacifica* productivity. We measured nitrogen fluxes using intact sediment cores collected from the lower, middle, and upper marsh positions during the wet (winter-spring) and dry (summer-fall) seasons to capture changes in N transformations, particularly N-removal. We found that changes in N transformations were associated with freshwater-driven belowground plant production across the marsh platform. Specifically, net N-removal rates were >2.5X higher in upper and middle marsh positions, which had increased belowground plant productivity due to fresh groundwater upwelling. We also found a strong seasonal effect on N fluxes. The marsh served as a N source in the wet season and a N sink in the dry season. Overall, terrestrial freshwater inputs can help predict patterns in N-

transformations within heterogeneous marsh systems and will be useful as climate change continues to affect the composition and persistence of tidal marshes.

Keywords: denitrification, *Salicornia pacifica*, pickleweed, terrestrial-marine interface, groundwater, tidal marsh

Introduction

Tidal salt marshes are coastal filters that serve as control points for nitrogen (N) removal at the terrestrial-marine interface (Bernhardt et al., 2017; Jordan et al., 2011). While specific marsh conditions have been shown to influence N processing (e.g., redox potential, temperature, substrate availability; George & Antoine, 1982; He et al., 2016; Koop-Jakobsen & Giblin, 2009), less is known about how terrestrially derived freshwater inputs may influence microbial N processing, particularly removal via microbial denitrification. Given that terrestrial freshwater inputs influence vegetation composition and porewater geochemistry (Moffett et al., 2012; Xie et al., 2020), it is likely that spatial and seasonal variation in terrestrial freshwater is an underlying driver of patterns in N removal. In addition to providing insight into drivers of N removal in salt marshes, understanding how terrestrial water inputs influence biogeochemical processing is critical as climate change and other human-driven disturbances continue to alter the timing and magnitude of freshwater delivery to coastal environments (J. Guimond & Tamborski, 2021).

The primary pathway for permanent N removal in salt marshes is denitrification, the microbially-mediated stepwise reduction of nitrate (NO_3^-) to dinitrogen (N_2) and nitrous oxide (N_2O) gases. Denitrification rates are limited in part by the availability of NO_3^-, which in salt marshes comes primarily from coupled nitrification-denitrification, anthropogenic sources, or coastal upwelling (Bowen et al., 2024). Additionally, heterotrophic denitrification requires a supply of labile organic carbon (M. R. Hamersley & Howes, 2003). Finally, denitrification can be suppressed by sulfide accumulation, either through direct toxicity or from inhibition of nitrification, which limits NO_3^- availability (Joye & Hollibaugh, 1995; Sørensen et al., 1980).

Marsh vegetation plays an important role in regulating denitrification capacity by modulating these drivers (Alldred & Baines, 2016; M. Hamersley & Howes, 2005; Koop-Jakobsen & Giblin, 2009). In the rhizosphere, radial oxygen loss promotes coupled nitrification-denitrification and reduces sulfide inhibition (Joye & Hollibaugh, 1995; Koop-Jakobsen & Giblin, 2009), and labile carbon (C) exudates and biomass serve as a substrate source to heterotrophic microbes (Dodla et al., 2008; Henry et al., 2008). Additionally, evapotranspiration can increase porewater salinity during periods of high productivity (Montalvo et al., 2024; Morris, 1995), and competition for nutrients via plant uptake can limit microbial N transformations (Kuzyakov & Xu, 2013; Ouyang et al., 2016).

While importance of marsh vegetation as a driver of N-removal is well established, less is known about the role of terrestrial freshwater inputs in driving denitrification. Much of our understanding of terrestrial groundwater flowpaths in marshes is centered around N-flux to and from marshes (Osgood, 2000; e.g., Osgood & Zieman, 1998), rather than within-marsh processing. Terrestrial freshwater can deliver allochthonous N to the marsh, alter salinity,

and drive redox conditions (J. A. Guimond et al., 2025; Krause et al., 2020; Xin et al., 2022). Additionally, terrestrial freshwater inputs also influence patterns in vegetation composition and productivity (Wilson and others 2015; Karberg and others 2018), which in turn drive denitrification rates (Ledford et al., 2020; Ooi et al., 2022). Thus, understanding how terrestrial freshwater inputs influence soil denitrification may improve our ability to understand when and where control points form in salt marshes.

Here, we present a study investigating how tidal freshwater inputs affect biogeochemical processes within a tidal marsh dominated by a single plant species (*Salicornia pacifica*), located in central California (Elkhorn Slough). The study transect spans approximately 25 m between the terrestrial uplands and the lower boundary of the salt marsh. Previous work at the site has shown an elevation-driven gradient in tidal inundation and freshwater inputs (Grande et al., 2022; Montalvo et al., 2024). At upper marsh positions, hydrologic dynamics are controlled by seasonal freshwater delivery from the uplands during the wet season, while lower marsh water level and hydrologic dynamics are controlled by tidal forcing year-round and minimally dampened by freshwater inputs (Montalvo et al., 2024). These differences in hydrologic forcings impact the timing and range of porewater salinity. Specifically, during the wet season, specific conductance is lower in the upper marsh compared to the middle and lower marsh (Montalvo et al., 2024). The differences in water sources also influence primary productivity in the marsh; namely, freshwater inputs promote earlier greening and greater aboveground plant production in the upper marsh (Montalvo et al., 2024). In this study, we sought to test our hypothesis that the highest rates of N removal (measured as N_2 flux) would occur where freshwater influence was the strongest (i.e., the upper marsh) due to increased plant productivity.

Methods

Site description

This study took place at a salt marsh at Elkhorn Slough, which is a shallow estuary (11 km long; 3.8 km^2 area) that empties into the Monterey Bay National Marine Sanctuary in central California. Elkhorn Slough is part of the NOAA National Estuarine Research Reserve (NERR) network. Agricultural practices dominate land use surrounding Elkhorn Slough, which has resulted in excess nutrient inputs to the estuary, creating eutrophic conditions in some areas (Jeppesen et al., 2018). The estuary receives a mixed semidiurnal tidal cycle (~2.7 m in range; Breaker et al., 2008; Grande et al., 2022), and its dominant surface freshwater source, the Salinas River,

enters Elkhorn Slough at its mouth. The region is characterized by a Mediterranean climate, with rainfall occurring predominantly in winter and spring, which is out of phase with the dominant growing season in summer.

The study marsh is located at Cowell Ranch (36°51'5.93"N, 121°45'44.62"W), approximately 9 km from the estuary's mouth (Fig. 1). This marsh study site lies along the intertidal zone with a local tidal range of -1.9 to 1.6 m above mean sea level (amsl). The dominant plant species in the marsh is pickleweed (*Sarcocornia pacifica*, also known as *Salicornia pacifica*), a perennial halophyte native to North America. *S. pacifica* productivity is highest during the summer growing season and lowest during senescence and dormancy in winter (Montalvo et al., 2024).

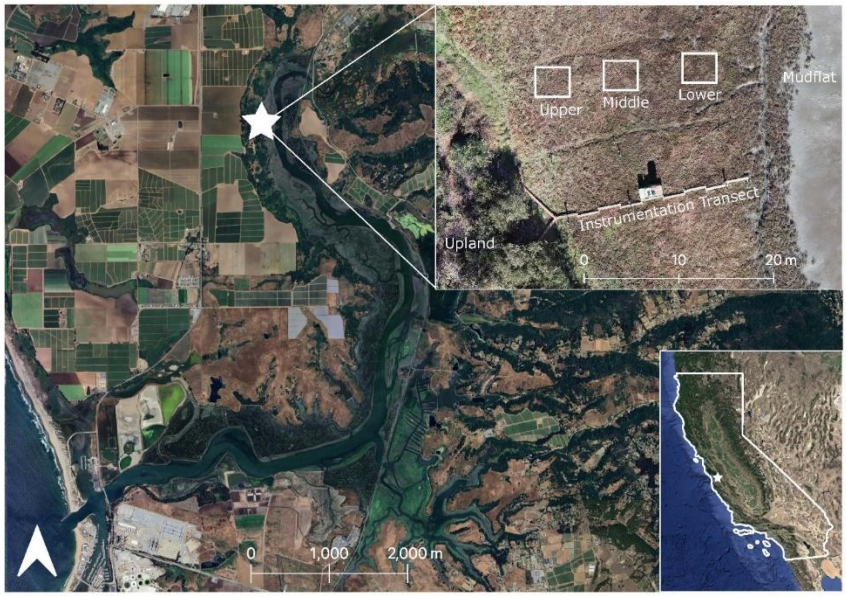


Fig. 1 A map of the study site (indicated by star). The sampling transect is shown in the upper left inset. The location of Elkhorn Slough NERR within California, USA, is indicated by the start in the lower right inset.

Experimental Design

We used elevation to delineate study plots by lower (0.64 m amsl), middle (0.71 amsl), and upper (0.76 m amsl) marsh positions along the marsh transect that are defined by differences in water source mixing and proximity to the terrestrial uplands (Grande et al., 2022; Fig. 1). The lower, middle, and upper marsh positions are inundated by tides approximately 9.6, 7.2, and 5.4 % of the time, respectively (Montalvo et al., 2024). The median fraction of porewater that originated as terrestrial freshwater (not estuarine surface water) in the wet season for the lower, middle, and upper marsh positions were 11, 10, and 36%, respectively (Grande, Visser, et al., 2023). The median fraction of porewater that originated as terrestrial freshwater in the dry season for the lower, middle, and upper marsh positions were 0, 2, and 17%, respectively (Grande, Visser, et al., 2023). To assess how terrestrial freshwater contributions affect salt marsh biogeochemical processes, we conducted incubation experiments on sediments

collected from each of these three marsh positions during different periods of plant activity across the wet and dry seasons. We conducted three experiments during the wet season (11 February 2021, 7 March 2021, and 8 April 2021), and three experiments during the dry season (19 November 2020, 6 May 2021, and 12 July 2021). Of these time points, February, March and November were during the dormant season, while April, May and July were during the growing season.

Water level monitoring

We installed a Solinst 3001 LT M5 pressure transducer (Solinst, Ontario, Canada) in a 2.5 m deep piezometer in the uplands, roughly 5 m upslope from the salt marsh boundary. We used water level data as a proxy for changes in water sources between the wet and dry seasons. Please read Montalvo and others (2024) Grande and others (2022) for more details on water level monitoring.

Surface water and porewater characterization

During each of the six experiments, we collected surface water samples at the site during high tide conditions or from Kirby Park, a boat launch approximately 2 km away across from our study site. We collected the surface water samples in triplicate, filtered (PES, 0.45 µm), and immediately froze for nutrient analysis. We collected triplicate porewater samples from 10 cm depth at each position using a plastic sipper and syringe, which were filtered (PES, 0.45 µm), and frozen for nutrient analysis. Collecting the samples at 10 cm allows us to capture porewaters within the rhizosphere without drawing down surface water. In the field, we measured porewater dissolved oxygen, temperature, pH, and electrical conductivity with a handheld water quality meter (Orion Star A321). Dissolved inorganic nitrogen (NH_4^+, NO_3^-, and NO_2^-) concentrations were measured in surface water and porewater samples on a Lachat Quikchem 8500 Autoanalyzer at the Marine Analytical Laboratory at the University of California, Santa Cruz, using a cadmium reduction column following the EPA 353.2 method (O'Dell, 1993). The instrument detection limit was 0.01 mg/L, verified with a series of calibration dilution tests.

Core incubation experiments

We determined denitrification rates by measuring dinitrogen (N_2) gas in intact sediment core incubations. For each experiment, we collected marsh sediments in acrylic cores (13 cm x 10.2 cm i.d.) in triplicate from each marsh position (n = 9 cores). We filled the core headspace with site water adjacent to the marsh platform and sealed for

transport to the lab on ice. Once in the lab, we submerged uncapped cores in site water with aquarium bubblers in a darkened environmental chamber at surface water temperature. We submerged triplicate cores in the same water bath, and we added a fourth "blank" core containing only site water to each bath to capture water column activity. Following an overnight incubation, we capped the cores with overlying water and set up a gravity flow system (Greene, 2020; Owens & Cornwell, 2016), which sent filtered (PES 0.45 μm) surface water into the cores. Overlying water was stirred with magnetic stir bars attached to the core caps that were controlled by a central motorized magnetic stirring unit (Greene, 2020). The stir speed of the magnets was low to avoid suspending sediment or producing bubbles in the headspace, and magnets were visually inspected to ensure comparable and consistent rotation speed across cores. Starting at the time of capping (t0), we collected outflow samples for dissolved gases (N_2, Ar) in Exetainer vials (Labco), preserved with zinc chloride (50% w/v) and stored in a temperature-controlled water bath. We filtered outflow samples (PES 0.45 μm) for dissolved inorganic N (NH_4^+, NO_3^-, NO_2^-) and froze for analysis. We collected dissolved gas and water samples at 3-hour intervals up to 12 hours, or until dissolved oxygen dropped below 10%. Gravity-fed surface water replaced the volume of extracted water from the headspace during sampling.

To determine N_2 fluxes across the sediment-water interface, we measured N_2 relative to Ar on a membrane inlet mass spectrometer (MIMS) outfitted with a copper reduction column (Bay Instruments) (Eyre et al., 2002; Kana et al., 1994). We calculated N_2 fluxes across the sediment-water interface by linear regression of N_2 production, corrected for dilution by replaced water, as a function of incubation time, core water volume, and surface area. We corrected these rates for water column activity by subtracting blank core rates from sediment core rates. Only linear portions of the incubation time curve were used, and if the regression was not significant ($p<0.05$), the flux was considered zero. Positive N_2 fluxes are indicative of N removal via denitrification. Negative fluxes are indicative by N production via N-fixation.

We measured dissolved NH_4^+ and NO_3^- on a Lachat Quikchem Autoanalyzer with a cadmium reduction column. We calculated fluxes for each N species using the same approach as for N_2. Positive NH_4^+ and NO_3^- fluxes are indicative of N production through mineralization or nitrification. Negative NH_4^+ and NO_3^- fluxes are indicative of N consumption.

Core sediment processing

Following the incubation experiment, we removed the top 5 cm of each core for bulk sediment C and N and to determine plant belowground biomass. To determine bulk C and N, we freeze dried and ground a representative 4-5 gram sediment subsample of the top 5 cm for elemental analysis using a CN-EA-iRMS in the Stable Isotope Laboratory at University of California Santa Cruz. We preserved the remaining sample to determine total belowground biomass by separating roots from the soil by wet sieving (2 mm mesh), then drying to a constant weight at 65 °C.

Vegetation Characterization

To characterize plant activity during the study period, we used a 0.5 m x 0.5 m quadrat to measure the percent cover of green (turgid), woody (brown, senesced) and bare area in two replicate plots at each marsh position (Montalvo et al., 2024). These plots were composed solely of *S. pacifica*. We determined plant belowground biomass from the preserved core subsamples, as described in the previous section.

Data analysis

To confirm that sampling dates were accurately categorized as wet or dry season, we ran principal components analysis (PCA) on key environmental conditions that vary seasonally. The PCA of air temperature, surface water temperature, porewater temperature, porewater conductivity, and terrestrial upland water level showed clear separation that correlated with wet and dry months described in the Experimental Design section above (Supplemental Material). Thus, experimental data were compared between wet and dry seasons, rather than across individual dates.

We analyzed nitrogen fluxes, plant belowground biomass, soil C:N, plant percent cover, plant percent live biomass, plant percent woody biomass, and porewater DIN using two-way analysis of variance (ANOVA) with season (wet or dry) and marsh position (lower, middle, upper) as factors. We tested significant post hoc effects using Tukey's Honestly Significant Difference (Tukey's HSD) test. We tested the assumptions of ANOVA by visual inspection of

236 residuals on a QQ plot and Levene's test. We analyzed surface water nutrients by season using Kruskal-Wallis tests.
237 When necessary, we transformed the data to meet the assumptions of ANOVA.
238
239 **Results**
240 *Surface and porewater DIN*
241 Surface water NO_3^- ranged from 0-37 µM and did not differ between seasons (Kruskal-Wallis, χ^2=1.25, df=1,
242 p=0.264). Surface water NH_4^+ ranged from 8-40 µM and was nearly 3 times higher during the wet season (27 ±12
243 µM) compared to the dry season (10 ±2.5 µM) (Kruskal-Wallis, χ^2=7.35, df=1, p=0.007).
244
245 Porewater NO_3^- concentrations ranged from below detection to 11.2 µM across the study period (Fig. 2a). One-third
246 of NO_3^- concentrations were below detection. There were no effects of season (ANOVA, df=1, F=0.06, p=0.803) or
247 position (ANOVA, df=2, F=2.52, p=0.094) on porewater NO_3^- concentrations. NH_4^+ was the dominant form of
248 porewater DIN, and was highly variable, ranging from 6.5 to 345 µM across the study period (Fig. 2b). Porewater
249 NH_4^+ was marginally higher during the dry season (100 ± 95 µM) compared to the Wet season (72 ±75 microM)
250 (ANOVA, df=1, F=3.07, p=0.088). Porewater NH_4^+ differed between marsh positions (ANOVA, df=2, F=58.99,
251 p<0.001), decreasing nearly 10-fold from the lower marsh to the upper marsh (Tukey's HSD, p<0.034).
252

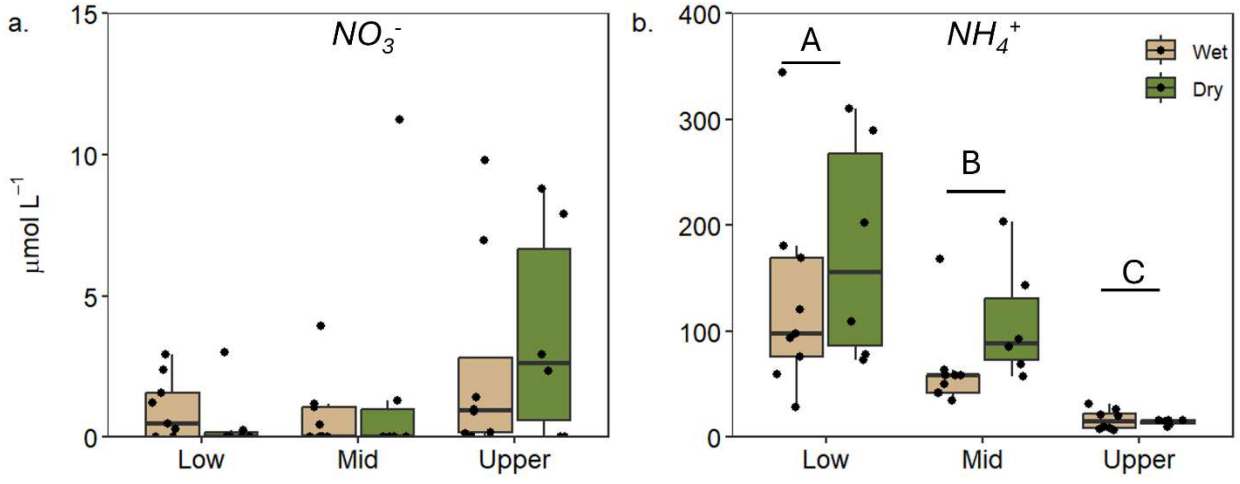


Fig. 2 Porewater concentrations of a. NO_3^- and b. NH_4^+ during the wet (tan) and dry (green) season for each marsh position. Note the difference in y-axes. The box indicates the median and interquartile ranges. Whiskers extend to highest and lowest points no more or less than 1.5 times the interquartile range. Dots indicate individual measurements. Letters indicate differences between marsh positions.

Nitrogen Flux rates

There were effects of marsh position (ANOVA, df=2, F=4.69, p=0.014) and season (ANOVA, df=1, F=10.06, p=0.003) on N_2 flux, though there was no interaction effect (ANOVA, df=2, F=0.33, p=0.723) (Fig. 3a). The lowest N_2 flux rates occurred in the lower marsh position across all seasons and were >2.5 times lower on average compared to the upper marsh position (Tukey's HSD, p = 0.017), and were marginally (Tukey's HSD, p=0.054) lower than the middle marsh position. There was no difference in N_2 flux rates between the upper and middle marsh positions (Tukey's HSD, p =0.887). N_2 flux rates also varied between seasons across all marsh positions. On average, N_2 flux rates were 2.3 times higher during the dry season compared to the wet season. All negative or zero flux measurements occurred during the wet season. More than half (52%) of NO_3^- flux rates were equal to zero; that is, there was no linear production or consumption of NO_3^- during the incubation period. There was no effect of marsh position on NO_3^- flux (ANOVA, df=2, F=1.09, p =0.343) (Fig. 3b). However, there were dramatic seasonal differences (ANOVA, df=1, F=7.08, p=0.004), with the marsh serving as a net NO_3^- sink during the dry season (-120 ± 198 [SD] μmol m^{-2} h^{-1}) and a net NO_3^- source during the wet season (430 ± 835 [SD] μmol m^{-2} h^{-1}).

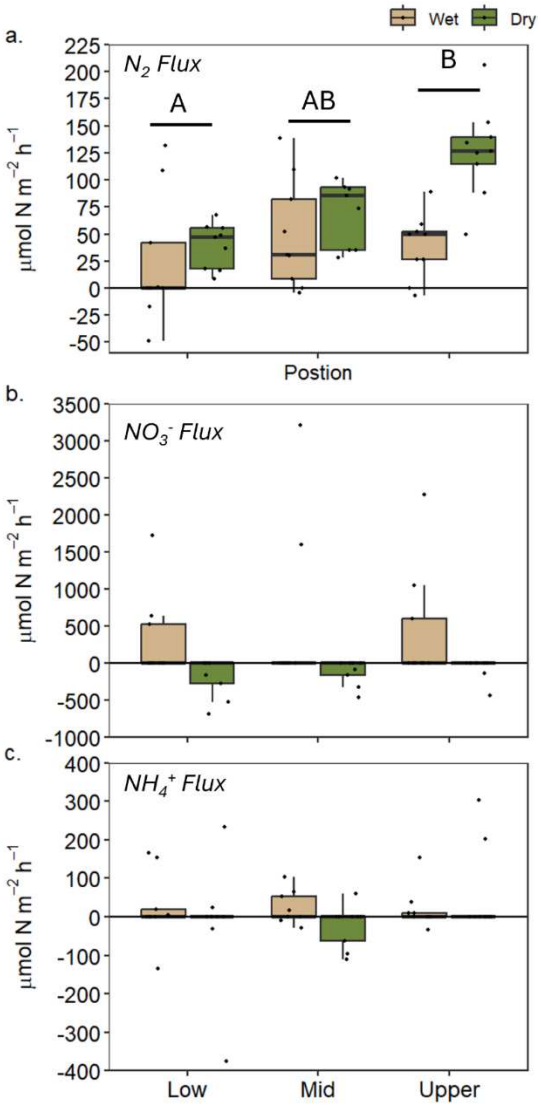


Fig. 3 Seasonal a. N_2, b. NO_3^-, and c. NH_4^+ fluxes across low, middle, and high marsh positions. Note the difference in y-axes. Letters indicate differences between marsh positions. Seasonal effects are not indicated—N_2 flux was higher and NO_3^- flux lower during the Dry season across all positions. The box indicates the median and interquartile ranges. Whiskers extend to highest and lowest points no more or less than 1.5 times the interquartile range. Dots indicate individual flux measurements.

As with NO_3^- flux, more than half (52%) of NH_4^+ flux rates were equal to zero. The majority (65%) of non-zero NH_4^+ flux rates were positive (Fig. 3c). There was no effect of marsh position (ANOVA, df=2, F=0.89, p=0.419) on NH_4^+ flux. The marsh was a net source of NH_4^+ during the dry (5 ± 117 [SD] μmol m^{-2} h^{-1}) and wet (22 ± 63 [SD] μmol m^{-2} h^{-1}) seasons, though the difference between seasons was marginal (ANOVA, df=1, F=0.93, p=0.061).

Soil C:N

Soil C:N varied across marsh positions (ANOVA, df=2, F=21.28, p=<0.001), increasing with marsh position (low: 13.3±2.7; middle: 14.9±0.8; upper: 16.7±0.9) (Fig. 4a). There was no effect of season on soil C:N (ANOVA, df=1, F=1.41, p=0.242).

Vegetation characterization

Percent cover of aboveground biomass ranged from 47.2% to 100%. There was no effect of marsh position (ANOVA, df=2, F= 0.90, p=0.4144) or Season (ANOVA, df=1, F=0.09, p=0.7724) on percent cover. There was no effect of marsh position on percent live biomass (ANOVA, df=2, 1.88, p=0.1642; Fig. 4b), but there was an effect of season (ANOVA, df=1, F=35.74, p<0.001). The percentage of live biomass was 3 times higher in the Dry season (51% ±24%) compared to the Wet season (17%±20% (Fig. 4b).

Unlike aboveground biomass metrics, plant belowground biomass differed between marsh positions (ANOVA, df=2, F=14.68, p<0.001) (Fig. 4b). The effect of position was dependent on season (Position x Season, df=2,

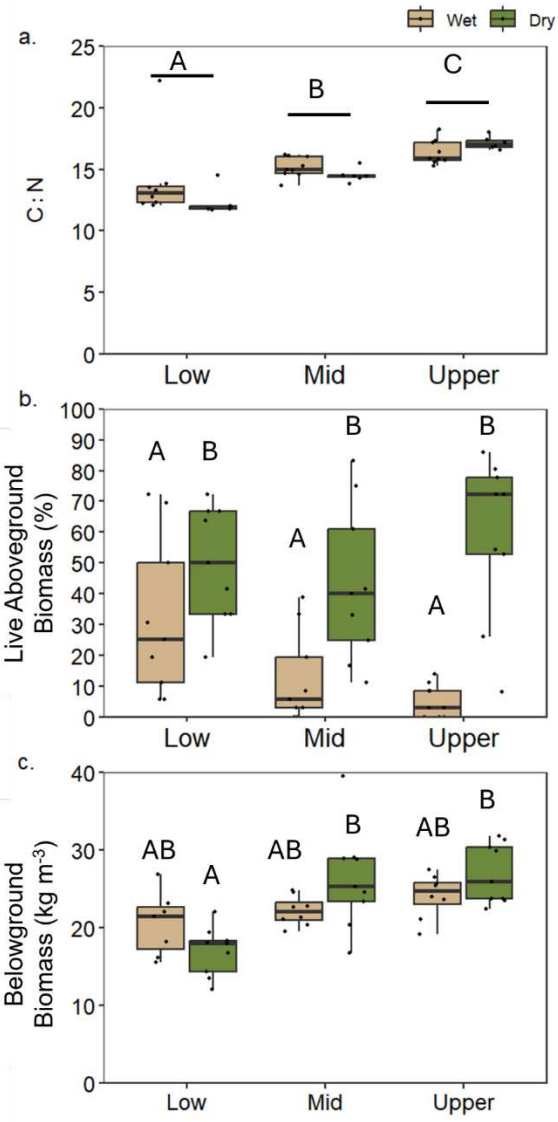


Fig. 4 Seasonal a. soil bulk carbon to nitrogen ratio, b. Percent live plant biomass, and c. belowground plant biomass. Letters indicate differences between marsh positions and seasons. The box indicates the median and interquartile ranges. Whiskers extend to highest and lowest points no more or less than 1.5 times the interquartile range. Dots indicate individual flux measurements.

F=4.26, p=0.020). During the Dry season, plant belowground biomass was lower in the low marsh (16.9±3.1 kg m^{-3}) than the middle (26.3±2.1 kg m^{-3}) or upper (26.9±3.8 kg m^{-3}) marsh positions (Tukey's HSD, $p<0.001$). In contrast, there was no difference between positions during the Wet season (Tukey's HSD, p=0.508), nor was there an overall difference in biomass between the Wet and Dry seasons (ANOVA, df=1, F=1.21, p=0.277).

Discussion

Terrestrial freshwater inputs influence marsh biogeochemical function by driving changes in plant productivity

Our findings indicate that nitrogen processing responds to changes in freshwater inputs that drive plant productivity. In a study at the same field site, Montalvo and others (2024) found that upper marsh positions were characterized by less saline porewater concentrations, which was specifically driven by greater terrestrial freshwater inputs, and this in turn was linked to earlier greening and higher aboveground production in the upper marsh. Similarly, this current study found that plant belowground biomass increased with elevation during the growing season, confirming that terrestrial freshwater inputs enhance both above and below ground plant productivity. We expect that hydrologically driven changes in plant productivity across the marsh platform drove spatial patterns in N_2 fluxes, serving as an ultimate controller of N-removal capacity. Historically, hydrologic impacts on marsh structure and function have been considered as a result of hydrologically-driven patterning of plant species distribution (e.g., Wilson et al., 2015), but our findings indicate that terrestrial freshwater inputs drive changes within a single plant species, resulting in spatiotemporal differences in biogeochemical functioning.

Much of our understanding regarding the impact of freshwater inputs on N removal in tidal marshes considers direct effects, such as changes in redox conditions, salt flushing, and electron acceptor delivery (Grande et al., 2022; Tobias et al., 2001; Xie et al., 2020). However, our findings indicate that terrestrial freshwater inputs indirectly mediated N removal through changes in plant productivity, rather than direct effects on edaphic conditions. Specifically, we found that freshwater upwelling in the upper marsh increased plant productivity, which was indicated by higher belowground biomass and soil C stocks, resulting in elevated N_2 fluxes (i.e., denitrification) compared to the tidally dominated low marsh. Plant belowground activity promotes N removal via denitrification through radial root oxygen loss (Koop-Jakobsen et al., 2018; Moffett & Gorelick, 2016), which can alleviate sulfide inhibition of denitrification (Joye & Hollibaugh, 1995; Sørensen et al., 1980). Additionally, root exudates provide a

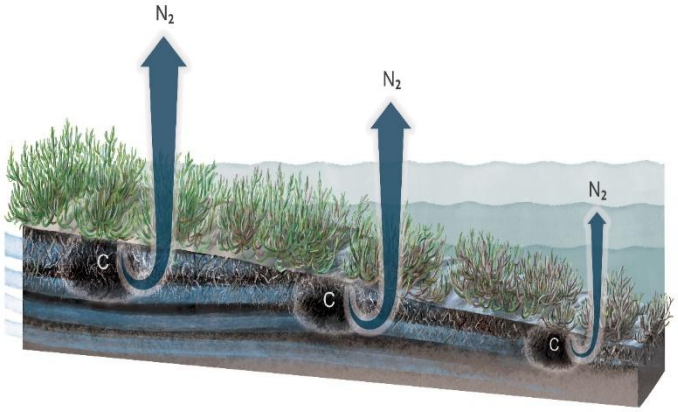


Fig. 5 Conceptual summary of functional dinitrogen gas flux at Elkhorn Slough. In the upper and middle marsh positions, terrestrial freshwater inputs (indicated by blue bands) support greater belowground plant productivity, increasing soil organic matter stocks. This results in higher nitrogen removal rates (i.e., positive N_2 flux; blue arrows) compared to the low marsh position, which is dominated by tidal water inputs.

334 labile C source that can promote denitrification (Henry et al., 2008; Spivak & Reeve, 2015). Parsing out variability within a species, even when differences in freshwater inputs are too subtle to result in species composition differences, can allow predictions to be made about differences in biogeochemical function based upon readily available hydrologic data. This is useful in low gradient tidal marshes, like Elkhorn Slough.

In addition to N-removal, our data supports the hypothesis that other N-processing rates differed due to terrestrial freshwater inputs. Specifically, we expect that lower belowground plant productivity and higher tidal contributions in the lower marsh position favored conditions for dissimilatory nitrate reduction to ammonium (DNRA) over denitrification, particularly during the dry season when higher evapotranspiration and less freshwater flushing increases rhizosphere salinity (Kessler et al., 2018). DNRA is a competing NO_3^- reduction process that is favored over denitrification when organic matter is high relative to NO_3^- and/or highly reducing conditions result in sulfide accumulation (Hardison et al., 2015; Murphy et al., 2020). This is supported by our flux data, as negative NO_3^- fluxes (i.e., NO_3^- consumption) were comparable across marsh positions whereas positive N_2 fluxes (i.e., N removal) were lower in the low marsh, indicating an alternate fate for NO_3^-. Additionally, DNRA can be a source of porewater NH_4^+ in tidal marshes (Giblin et al., 2013, p. 201), and we measured higher porewater NH_4^+ concentrations in the low marsh. While more direct measurements of DNRA are necessary to confirm our hypothesis, our findings suggest that terrestrial freshwater inputs may be an important driver of the fate of NO_3^- in tidal marshes.

Hydrologically mediated changes in plant productivity drove spatial patterns in biogeochemical processing, but temporal patterns were driven by seasonal freshwater inputs

While differences in plant productivity across the marsh platform resulted in spatial variation in N removal rates, seasonal drivers, specifically the magnitude of freshwater inputs, were more important in determining whether the marsh was a net source or sink for N. Both N_2 and DIN fluxes followed a general pattern of the marsh being a DIN source during the wet season and a DIN sink during the dry season, across all marsh positions. This finding is consistent with previous research at the site, which found that the marsh is a net source of NO_3^- during the wet season, and net sink of NO_3^- in the dry season (Grande, Seybold, et al., 2023). We expect that this is due to a mismatch in the timing of maximum NO_3^- delivery and peak microbial activity. Specifically, winter precipitation events and tidal inundation deliver DIN to the marsh platform during the wet season (Grande, Seybold, et al., 2023), coincident with lower denitrification rates. We expect that this reduction in denitrification rates is caused by plant dormancy (Montalvo et al., 2024) and colder temperatures (George & Antoine, 1982; Pfenning & McMahon, 1996), leading to the salt marsh filtering less terrestrial N during the wet season.

Despite measuring positive DIN fluxes during the wet season incubations, we observed limited changes in *in situ* porewater NO_3^- concentrations. Similarly, Grande and others (2023) found that absolute changes in NO_3^- concentrations between wet and dry seasons were relatively small. It is possible that *in situ* porewater NO_3^- concentrations reflect plant uptake during the wet season, which would not be reflected in core incubations, which measure microbial activity. The vegetation at Elkhorn Slough remains N limited despite receiving N loads and has a high capacity for N uptake even during the wet season (Nelson & Zavaleta, 2012).

Conclusions & Implications

We found that lower plant belowground productivity in the more tidally influenced low marsh position likely contributed to lower N-removal rates than the middle and upper marsh positions. As sea levels continue to rise, we expect that increased inundation and salinity stress will reduce plant productivity at higher marsh elevations, resulting in a functional shift where a larger portion of the marsh behaves functionally like the low marsh. The interaction of terrestrial freshwater and tidal saltwater intrusion impact plant composition and health in marsh-upland transition zones (Pratt et al., 2026), which in turn influence marsh persistence and function in response to sea level rise. Marsh migration into uplands provides a mechanism for marsh persistence in the face of sea level rise

(Kirwan, Temmerman, et al., 2016). At Elkhorn Slough, increased tidal inundation from sea level rise has resulted in rapid inland movement of the marsh-upland ecotone boundary (Wasson et al., 2013). As sea levels continue to rise, we expect that freshwater-supported areas within the marsh will become increasingly significant control points for N-removal. These areas may be of particular significance as marsh landward migration on the US Pacific coast is limited by topography (Kirwan, Walters, et al., 2016) – by supporting biogeochemical control points at the marsh-upland ecotone, freshwater upwelling zones can facilitate marsh migration into uplands.

Climate change poses the additional challenge of altering terrestrial freshwater inputs into marshes, which in turn can disrupt biogeochemical processes. In central California, climate forecasts predict limited change to total annual precipitation, but more intense precipitation events that are less frequent (Swain et al., 2018). This intensifying precipitation regime may limit the potential for groundwater recharge, which may diminish seasonal groundwater inputs to tidal marshes. This, in turn, may limit the extent of freshwater-driven N-processing. Or, a reduction in freshwater inputs may shift the dominant pathways of N-processing, such as promotion of DNRA over denitrification. Ultimately, the impacts of climate change on marsh hydrology and functional capacity are complicated, as the interactions between sea level rise and precipitation change will alter both the extent and timing of plant and microbial activity within tidal marshes.

Ethics approval and consent to participate: This research does not involve human or animal subjects.

Data Availability: Data will be made publicly available via ESS-DIVE, with the link and DOI provided for the final publication.

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