Submarine groundwater discharge: Nitrogen biogeochemistry of the discharge zone

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Abstract

To investigate the role of the seepage zone in transport, chemical speciation, and attenuation of nitrogen loads carried by submarine groundwater discharge, we collected near-shore groundwater samples (n=328) and examined distribution and isotopic signature ($\delta^{15}N$) of nitrate and ammonium. In addition, we estimated nutrient fluxes from terrestrial and marine-source groundwater. We discuss our results in the context of three aquifer zones: a fresh groundwater zone, a shallow salinity transition zone (STZ), and a deep STZ. Groundwater plumes containing nitrate and ammonium occurred in the freshwater zone, while the deep STZ carried almost exclusively ammonium. The distributions of redox-cycled elements were consistent with theoretical thermodynamic stability of chemical species, with sharp interfaces between water masses of distinct oxidation/reduction potential, suggesting that microbial transformations of nitrogen are rapid relative to dispersive mixing. In limited locations where overlap occurs between distribution of nitrate with that of ammonium and dissolved Fe^{2+}, changes in concentration and in $\delta^{15}N$ suggest loss of all species. Mechanisms for concurrent removal of NO_3^- and NH_4^+, both in freshwater and the deep STZ, could be any of heterotrophic or autotrophic denitrification, coupled nitrfication/denitrification, anammox, or Mn oxidation of NH_4^+. Loss of nitrogen was not apparent in the shallow STZ, perhaps due to short water residence time. Despite organic C-poor conditions, the near-shore aquifer and subterranean estuary are biogeochemically active zones, where attenuation of N loads can occur. Extent of attenuation is controlled by degree of mixing of biogeochemically dissimilar water masses, highlighting the critical role of hydrogeology in N biogeochemistry. Mixing is related in part to thinning of the freshwater lens prior to discharge, and to dispersion at the fresh/saline groundwater interface, features common to all SGD zones.

Introduction

Eutrophication of coastal waters due to non-point source land-derived nitrogen (N) loads is a worldwide phenomenon (NRC 2000, Howarth et al. 2000). Within the United States, a majority of estuaries have been determined to be moderately to severely impaired by eutrophication associated with increasing nutrient loads (Bricker et al. 1999). In coastal watersheds with soils of high hydraulic conductivity and permeable coastal sediments, groundwater is commonly a major route of transport from land to sea for freshwater and associated land-derived nutrient loads (Valiela et al. 2000). A portion of the freshwater flowing down-gradient from coastal aquifers discharges directly to coastal waters through a seepage face that may be located near the intertidal zone or farther offshore (Giblin and Gaines 1990, Bokuniewicz 1992, Cable et al. 1997). Such direct discharge of groundwater into estuaries or the sea is referred to as submarine groundwater discharge (SGD). On a global scale SGD can rival riverine transport of chemical loads to sea, and on a regional scale it can equal or exceed riverine input of freshwater and/or solutes (Taniguchi et al. 2002, Slomp and Van Cappellin 2004, Kroeger et al. 2007).

In addition to the terrestrially-derived fresh groundwater and solutes, saline and brackish groundwater are often important components of SGD (Li et al. 1999, Taniguchi et al. 2002, Michael et al. 2005). Processes including tidal pumping, wave set-up, and dispersion along the boundary between discharging fresh groundwater and the saline groundwater wedge beneath, result in entrainment of saline groundwater, producing a gradient in groundwater salinity and resulting in discharge of brackish and saline groundwater. Hence, SGD often consists of a substantial amount of re-circulating seawater, and thus it might produce important fluxes of sediment-regenerated nutrients and other porewater materials to coastal waters. The salinity transition zone in near-shore aquifers has been referred to as a subterranean estuary (Moore 1999), and mixing of fresh and saline groundwaters in that zone, in the context of coastal sediments, may alter the chemical composition of the discharging fluid (Moore 1999, Slomp and Van Cappellen 2004).

With regard to the study of biogeochemical cycles and chemical loads to coastal waters, the near-shore aquifer, seepage face and subterranean estuary are under-studied zones in the aquatic cascade from watershed to sea. High potential rates of nitrate reduction processes (denitrification and dissimilatory nitrate reduction to ammonium) have been demonstrated to occur at locations where nitrate-bearing fresh groundwater contacts soils within or just beneath fringing salt marsh (Tobias et al. 2001, Addy et al. 2005). In many cases, however, flow paths carrying groundwater to estuaries or directly to sea bypass fringing salt marshes, and instead discharge occurs through permeable intertidal or subtidal sediments (Bohlke and Denver 1995, Nowicki et al. 1999). The influence of fringing wetlands on nitrogen delivery to coastal waters has been further diminished by widespread removal of those wetlands due to human activities (Bertness et al. 2002). Where rapid discharge of fresh, oxygenated groundwater occurs through coarse intertidal sediments, N transformations in the top m of sediment are minimal, at least in some locations (Nowicki et al. 1999, Giblin and Gaines 1990). However, there has been insufficient detailed examination of the biogeochemical conditions and behavior of nutrients in deeper portions of near-shore aquifers and in the mixing zones between fresh and saline groundwater. Depending on the biogeochemical setting, a range of N transformations may occur in that zone, including removal of fixed N due to N gas-producing microbial processes, resulting in attenuation of land-derived N loads.

A fundamental problem in evaluating the importance of groundwater discharge in marine geochemical budgets is the difficulty of collecting samples across the salinity gradients of coastal aquifers. We are using a stainless steel drive point piezometer system to collect groundwater/pore water samples in high-resolution depth profiles at the head of Waquoit Bay, MA, and analyzing a suite of tracers and biogeochemically cycled elements (e.g. Testa et al. 2002, Talbot et al. 2003, Abraham et al. 2003, Charette and Sholkovitz 2006). By focusing on an accessible and well-defined subterranean estuary, in unprecedented detail, our study provides an in-depth view of the major biogeochemical reactions operating in estuarine permeable sediments with submarine groundwater discharge.

Here we present an examination of nitrogen biogeochemistry based on distributions of nitrogen concentrations and natural abundance stable isotope ratios in near-shore fresh, brackish and saline groundwater. In addition, based on results of hydrological, radiochemical tracer, and seepage meter studies (Michael et al. 2005, Mulligan and Charette 2006) we separately estimate N fluxes from terrestrial and marine-source SGD. Although dissolved organic nitrogen (DON) can be quantitatively important in both fresh and saline groundwater (Kroeger et al. 2006, 2006a, 2007, Burnett et al. 2007), at the present study site we have not yet examined the role of DON, and present here information on cycling and loading of dissolved inorganic nitrogen (DIN).

Methods

Site Description—The study area at the head of Waquoit Bay is located in western Cape Cod, Massachusetts (Fig. 1). The Cape Cod aquifer is unconfined and is composed of unconsolidated, quartz and feldspar sand and gravel (Oldale 1992). Soils are of glacial origin and are mostly sandy loam. The sandy soils allow rapid percolation of rainwater into the aquifer, and ground water recharge followed by discharge to small streams or to estuaries accounts for nearly all of net precipitation to Cape Cod watersheds. Typical groundwater velocity on Cape Cod ranges from 110 to 365 m per year (LeBlanc et al. 1988), and annual rainfall averages 1130 mm (Lajtha et al. 1995). Atmospheric deposition of TDN, wet plus dry, is 12 to 15 kg N ha^{-1} yr^{-1} (Valiela et al. 1997, Bowen and Valiela 2001). Naturally vegetated areas range from grass and shrublands to pitch pine and mixed oak forests (Lajtha et al. 1995). The head of Waquoit Bay watershed has a human population density of 190 km^{-2}, and wastewater disposal is through on-site septic systems (Kroeger et al. 2006). At our study site in the near-shore aquifer, solid phase organic carbon is low (non-detectable to 0.075%) (Charette et al. 2005), and dissolved organic carbon concentrations range from 30 to 700 μmol L^{-1} (Charette and Sholkovitz 2006).

Field sampling and analyses—Results presented here primarily focus on groundwater samples collected in a series of depth profiles in a 20-m shore-perpendicular transect and collected between March 25 and April 6, 2003 (Fig. 1; transect A, piezometers 7, 6, 11, 3, 5, 12). We also discuss results from collections conducted June 7 to July 3, 2002, both along transect A and along a 180-m shore-parallel transect (Fig. 1; transect B, piezometers 1, 2, 3, 4, 9), and from collections conducted June 25 to July 8, 2003, along a 12-m shore-perpendicular transect west of transect A (Fig. 1; transect C, piezometers 13 to 16). Sampling was designed to produce snapshots of biogeochemical conditions across the fresh, brackish and saline groundwater zones within the region of groundwater discharge.

We collected groundwater samples using a stainless steel, drive-point, Retract-A-Tip piezometer (AMS, Inc. Idaho, USA; Charette and Allen 2006). Samples were brought to the surface through acid-washed plastic tubing using a peristaltic pump. At each sampling depth, several void volumes were pumped and then flow was passed through a flow-though cell with a YSI 600R multi-probe inserted. When readings stabilized for salinity, dissolved oxygen concentration (D.O.), oxidation/reduction potential, pH, and temperature, we recorded the values and then collected samples. Oxidation/reduction potential readings were converted to Eh (mV) units by adding 200 to the values. Eh units can be further converted to units of pε (pε at 25° C = Eh/1000 x 16.9) for comparison to ORP readings in similar studies (e.g. Kroeger et al. 2007). Samples for nutrient concentrations and natural abundance stable isotope ratios of nitrate and ammonium were passed through a Millipore polyethersulfone cartridge filter (0.45 µm pore size), into acid-washed polyethylene sample bottles, and stored on ice until returned to the laboratory. A replicate nutrient sample for PO_4^{3-} analysis was acidified to ~ pH 2 with 8 N sulfurous acid to prevent scavenging by precipitation of iron oxides. Samples were analyzed immediately or were frozen until analysis. Nutrient concentrations (NO_3^-+NO_2^-, PO_4^{3-}, NH_4^+, SiO_4^-) were analyzed by colorimetric techniques using a Lachat QuickChem 8000 nutrient autoanalyzer. Nitrate and nitrite were not separately quantified, and in this paper their sum is referred to as "nitrate". Ammonium and NO_3^- were isolated for analysis of their N stable isotope ratios using adaptations of the ammonium diffusion method from Holmes et al. (1998) and Sigman et al. (1997), respectively. The acid-trapped samples were analyzed at the University of California, Davis Stable Isotope Facility using a Europa Scientific Hydra 20/20 isotope ratio mass spectrometer. The ratio is expressed as $\delta^{15}N$ (‰) = $[(R_{\text{sample}} – R_{\text{reference}})/ R_{\text{reference}}] * 1000$, where R is $^{15}N/^{14}N$ and the reference is atmospheric N_2. Standard deviation of replicate analyses is typically 0.1 ‰. Dissolved ferrous iron (Fe^{2+}) was measured by the ferrozine colorimeteric method (Stookey 1970). Dissolved Mn concentrations in groundwater were measured using ICP-MS after dilution with Milli-Q water.

Groundwater and nitrogen fluxes—We estimated fluxes of nitrate and ammonium, separately from terrestrial and marine groundwater discharges, based on concentrations within those water masses and on groundwater discharge estimates from Mulligan and Charette (2006) and Michael et al. (2005). Briefly, Mulligan and Charette used multiple approaches to estimate fresh, saline, and total (fresh + saline) groundwater discharge. They estimated fresh groundwater discharge along the 610 m shoreline at the head of Waquoit Bay using Darcy's Law calculations and based on repeated measurements of hydraulic gradients near the discharge zone over a two-year period. The Darcy calculations agreed well with estimates based on a watershed water budget. Saline and total groundwater discharges were estimated based on radium and radon inventories, respectively, in the near-shore estuarine surface waters and on a high density of measurements of activities of those radiochemical tracers in the near-shore aquifer. A second estimate of saline discharge rate was made based on the difference between total groundwater discharge rate (radon inventory) and the estimated fresh groundwater discharge rate. Extensive seepage meter studies by Michael et al. (2005) indicated larger saline SGD fluxes offshore of the zones of fresh and brackish groundwater discharge that might not have been reflected in the very near-shore radium and radon inventories measured by Mulligan and Charette, and we took the seepage meter measurements as an upper estimate for saline SGD.

Results and Discussion

Biogeochemical setting—There were three primary zones within the aquifer based on salinity (Fig 2A): the fresh groundwater zone, the shallow salinity transition zone (STZ) at the shallow intertidal portion of the freshwater aquifer, and the deep STZ beneath the freshwater aquifer. The three zones were distinct in terms of both water and N sources, and in terms of biogeochemical setting and N chemistry. There may also be large differences in groundwater velocity, rate (volume per unit time) and timing of water fluxes through each of the zones (Michael et al. 2005). Because each zone is distinct, to make accurate calculations of SGD-driven fluxes and transformations and sources of materials, ultimately it will be critical to examine each zone separately. Such detailed study of concentrations and chemistry in each zone is uncommon in the literature. Therefore, below we first describe each zone before moving on to discussions of nitrogen transformations.

Freshwater aquifer—Within the sampled portion of the freshwater aquifer, D.O. concentration and oxidation/reduction potential (Eh) were generally relatively high west of transect A (see transect C in Web Appendix http://www.aslo.org/lo/toc/volXXXX.pdf) and low at transect A and eastward (Web Appendix). A plume of reducing groundwater, characterized by low Eh, low D.O. concentration, and high dissolved iron concentration (Testa et al. 2002, Charette and Sholkovitz 2006), occurred on the east end of the head of Waquoit Bay. The most prominent feature in terms of nitrogen concentrations in the near shore fresh aquifer was occurrence of a groundwater plume containing high concentrations of NH_4^+ and 2 plumes containing relatively high nitrate concentrations (Fig. 2B and 2C). The nitrate and ammonium plumes were constant features during 2 years of sampling. The nitrate plume was present along the entire 180 m wide shore-parallel transect (transect B), whereas the ammonium plume was present only at transect A and eastward. The ammonium plume co-occurred with the plume of relatively reducing groundwater with high dissolved iron concentration (Web Appendix).

We cannot make definitive statements regarding the sources for the nitrate and ammonium plumes in the freshwater zone, but examination of natural abundance N stable isotopic ratios (Web Appendix), and of geographic features within the watershed, do allow some speculation. Nitrate occurring at < 50 µmol L^{-1} at the two shallowest sampling locations in piezometer 7 in transect A likely had a nearby atmospheric deposition/soil source or fertilizer source based on shallow occurrence and on low $\delta^{15}N\text{-}NO_3^-$ of 2.1 ‰. The deeper nitrate and ammonium plumes have no obvious anthropogenic sources within the watershed. They are wider (> 180 m for the nitrate plume and > 60 m for the ammonium plume) than would be expected for a wastewater plume from an individual septic system, although mingling of multiple wastewater plumes due to dispersion within the aquifer could produce the observed pattern. Two additional possible sources for the deeper nitrate and ammonium plumes are 1) recharge to the aquifer from two freshwater ponds (Bog Pond and Bourne Pond) located in the watershed approximately 300 and 450 m north of Waquoit Bay (Fig. 1), and 2) displacement of adsorbed N in saturated or unsaturated sediments in the watershed by ion exchange associated with occurrence of sea salt in the freshwater aquifer. The nitrate and ammonium plumes approximately co-occur with salinity anomalies of 1 to 2 in the freshwater aquifer, and ratios of major ions within the salinity anomaly suggests that their source is likely sea salt (not shown). Two potential sources for the salinity anomalies and their association with the N plumes are: 1) on shore winds, particularly associated with storms, can carry sea spray and associated sea salts inland as far as several km (Valiela et al. 1996), resulting in displacement of adsorbed ammonium from soils by cation exchange (Duckworth and Cresser 1991, Valiela et al. 1996), or 2) the salinity anomalies may be associated with mixing of water masses in the nearshore aquifer, and may be extensions of the shallow and deep STZ associated with movements of freshwater/saltwater interfaces due to seasonal and inter-annual variations in aquifer recharge (Michael et al. 2005). We should mention also that the plumes of groundwater containing elevated concentrations of ammonium and nitrate might have a common source, and represent the reduced core and oxidized edges of the same plume. Regardless of their source, the nitrate and ammonium plumes allow us to observe the behavior of nitrogen in the near-shore aquifer, as we will discuss in the section “Biogeochemical Transformations”.

Shallow salinity transition zone—The shallow STZ (Fig. 2A) occurs within or near the region of fresh groundwater discharge and likely results from gravitational convection and tide and wave-induced mixing of saline surface water into the discharging portion of the freshwater aquifer (Bokuniewicz et al. 2004, Robinson et al. 2006). Modeling and seepage meter studies suggest that velocity of groundwater discharge is relatively rapid and that flow-paths are short in the shallow STZ (Michael et al. 2005, Robinson et al. 2006). The salinity contour (Fig. 2A) suggests that much of the freshwater aquifer discharges to the intertidal zone and without mixing into the deep STZ, but that just prior to discharge the fresh groundwater does mix to a significant degree with saline porewater in shallow beach sediment. Thus, much of the fresh groundwater ultimately discharges as brackish water.

The saline water that mixed into the shallow STZ was likely very recently recharged, and commonly had high D.O. concentration and high Eh (Table 1, Fig 2). In both transects A and C, the shallower nitrate plume in the freshwater aquifer appeared to mix into the shallow STZ prior to discharge (compare Fig. 2A and 2B), so that nitrate concentrations in that zone were often quite high, while ammonium concentrations were relatively low (Table 1, Fig. 2C). As we will discuss further, at piezometer 4, east of transect A, mingling of the freshwater nitrate and ammonium plumes occurred within the shallow STZ.

Deep salinity transition zone—The deep STZ occurs at the base of the freshwater aquifer due to dispersive mixing of the discharging aquifer with the wedge of saline groundwater beneath (Fig. 2A). The occurrence of salinity lower than that of overlying bay water (28 to 29 ppt) suggests that mixing of the fresh and saline water masses occurs to as deep as ~8 m within the sediments. Modeling (Li et al. 1999, Michael et al. 2005), seepage meter (Michael et al. 2005) and radiochemical tracer (Abraham et al. 2003, Mulligan and Charette 2006) studies suggest that entrainment of saline porewaters by discharging fresh groundwater drives a brackish and saline groundwater circulation cell, and results in discharge of brackish and saline groundwater just offshore of the freshwater discharge maximum.

The higher salinity portions of the deep STZ are relatively reducing in comparison to the freshwater and brackish zones (Web Appendix and Table 1). Sulfide concentrations were not measured in this study, but within the deep STZ and saline porewaters sulfide odor was detectable only occasionally and only in shallowest (15 to 30 cm below sediment surface) porewaters at highest salinity. Alhough lack of sulfide odor does not preclude possibility of sulfate reduction followed by rapid precipitation as metal sulfides, measured oxidation/reduction potentials (Web Appendix and Table 1) suggest that reduced sulfur should not be thermodynamically stable within the deep STZ (Stumm and Morgan 1996). In addition, Charette and Sholkovitz (2006) did not observe significant sulfate depletion within the deep STZ. The presence of high concentrations of reduced N, Mn and Fe in solution suggest that those elements are important electron acceptors in much of the deep STZ. In the deepest and highest salinity portions of the deep STZ elevated concentrations of dissolved, reduced Fe occur, while at shallower depths and at lower salinity a Mn reduction zone occurs (Testa et al. 2002, Charette and Sholkovitz 2006).

Within the deep STZ, nitrate is largely absent (Fig. 2B) while ammonium is ubiquitous and is the dominant form of dissolved N (Fig. 2C, Table 2). The source for the ammonium in the deep STZ is likely remineralization of organic nitrogen in saline estuarine sediments, based on highest concentrations at or near maximum salinity, followed by approximately conservative mixing down the salinity gradient (Fig 3). Apparent conservative transport of ammonium within the deep STZ suggests that ammonium concentration is controlled to a large extent by the balance between production by remineralization, primarily at shallow depth in the estuarine sediment, and dilution by advecting groundwater. However, as we will discuss in the following section, N transformations do occur in locations where terrestrial nitrate mixes into the deep STZ. We have not determined here the extent of any coupled nitrification/denitrification that may occur at the site of remineralization in the saline groundwater recharge zone.

Biogeochemical transformations—To further examine behavior of N in the coastal aquifer, we measured natural abundance stable isotope ratios ($\delta^{15}N$) of nitrate and ammonium in a subset of the samples collected (Fig. 4). In broad terms the stable isotope data suggest the following: in the freshwater zone, nitrate shows a clear pattern of increasing $\delta^{15}N$ with decreasing concentration (Fig 4A). Such a pattern is indicative of loss of nitrate by an isotopically fractionating process, such as microbial reduction to N_2 or other gaseous form, in which the lighter isotope reacts more rapidly than does the heavier isotope, so that the residual material is enriched in the heavier isotope (Mariotti et al. 1981). Nitrate in the shallow STZ shows no clear indication of microbial transformation, with $\delta^{15}N$ remaining approximately constant with respect to concentration. Since occurrence of nitrate in the deep STZ is uncommon, we have only 2 measurements of $\delta^{15}N$, and both are at low concentration and relatively elevated $\delta^{15}N$ (Fig. 4A). $\delta^{15}N$ of NH_4^+ in the freshwater zone shows a tendency in some locations to increase with decreasing concentration, again suggesting NH_4^+ loss by an isotopically fractionating process (Fig. 4B). There may be some indication of NH_4^+ loss in the shallow STZ, with general increase in $\delta^{15}N$ with decrease in concentration, though the pattern is not consistent. $\delta^{15}N$ of NH_4^+ in the deep STZ shows a clear pattern of increase with decreasing concentration, consistent with loss of NH_4^+ in that zone (Fig. 4B).

Co-occurrence of nitrate and ammonium is uncommon throughout the near-shore aquifer and subterranean estuary, with both N forms occurring at concentrations > 2 µmol L^{-1} in only 19 of 328 samples collected (Fig. 5). Analysis of the natural abundance stable N isotope ratios along transect A suggests that the largely mutually exclusive distributions of nitrate and ammonium are in part due to loss of both N species in locations, both in the freshwater aquifer and deep STZ, where converging flow-paths bring nitrate-bearing waters with low D.O. concentration into close proximity with relatively reducing ammonium-rich waters, followed by dispersive mixing of the water masses. In those mixing zones, both nitrate and ammonium exhibit concurrent decrease in concentration and increase in N stable isotopic ratio (Fig. 6A to 6F, shaded regions). Co-occurring with, or often preceding, decrease in NH_4^+ concentration is decrease in concentration of dissolved Fe^{2+}, so that distributions of Fe^{2+} and NO_3^- rarely overlap (Fig. 6A to 6F). That pattern suggests concurrent loss of ammonium and nitrate, perhaps due to denitrification, coupled nitrification/denitrification or to other N_2 producing microbial processes such as anammox. The pattern of N loss shown in Fig. 6 is a consistent one, observed in both the freshwater zone (shaded regions in Fig. 6A to C and at 3 m depth in Fig. 6D to F) and deep STZ (shaded region at 5.5 m depth in Fig. 6D to F) portions of the near-shore aquifer.

Mixing of the nitrate and ammonium plumes in the freshwater zone is likely related to thinning of the aquifer and change in flow direction near the seepage face, so that the nitrate and ammonium plumes are brought into closer proximity. In addition, greater dispersion may be expected near the seepage face due to movement of water masses in response to tides and to seasonal cycles of recharge to the fresh aquifer. The shallower and deeper nitrate plumes, and the ammonium plume that occurs between the two nitrate plumes, converge just prior to discharge to the estuary (Fig. 2). The apparent result is that the ammonium plume is almost entirely consumed prior to discharge (Fig. 2C). The nitrate plumes also diminish substantially in concentration (Fig. 2B). In contrast to those N losses observed at transect A, at transect C, where the nitrate plume occurs in the absence of an ammonium plume and in groundwater with generally greater D.O. concentration, the nitrate plume appears to discharge without attenuation (Web Appendix; Talbot et al. 2003), further suggesting that mixing of the reduced and oxidized groundwater plumes at transect A is responsible for the fixed N loss prior to discharge.

In the deep STZ, although ammonium concentration appears to mix roughly conservatively from high concentration in saline groundwater to zero concentration in fresh groundwater (Fig 3), a similar plot including $\delta^{15}N\text{-}NH_4^+$ suggests that ammonium loss does occur in the few locations where we were able to observe small concentrations of terrestrial NO_3^- mixing into the deep STZ (Fig. 7). Limited measurements of $\delta^{15}N\text{-}NO_3^-$ and $\delta^{15}N\text{-}NH_4^+$ in the shallow STZ suggest that significant N transformations do not likely occur in the examined portions of that zone. In summer 2002, we sampled a location in which mixing occurred in the shallow STZ between the freshwater plume containing high ammonium concentration and the freshwater plume containing high nitrate concentration (Fig. 6G to I). In the mixing zone (shaded area), D.O. concentration was low (0.8 to 1.4 mg L^{-1}), and yet the N concentration and stable isotope ratios remain relatively low and do not show clear indication of loss of NO_3^- or NH_4^+. There may be some indication of nitrification, resulting in 15N enrichment of residual NH_4^+ pool and production of isotopically light NO_3^-. This is the only location sampled in which mixing of nitrate and ammonium-bearing groundwater did not result in substantial changes in concentration and $\delta^{15}N$. Such a result is likely related to the relatively rapid rate of groundwater flow and short flow-paths in the shallow STZ (Michael et al. 2005, Robinson et al. 2006), suggesting that mixing in the sampled location was recent, and that groundwater discharges to the estuary more rapidly than N transformations occur. Similar lack of N transformations during groundwater discharge has been observed previously in locations with rapid discharge of oxic groundwater (Giblin and Gaines 1990, Nowicki et al. 1999).

Estimates of nitrogen loss—To estimate the extent of N loss in the regions of overlap between nitrate and ammonium distributions in the freshwater zone and in the deep STZ, we used calculations based on the Rayleigh distillation model (Mariotti et al. 1981). In addition, we estimated the rate of N loss in the freshwater aquifer based on estimated rate of groundwater flow and on change in DIN inventory during transit to the discharge zone. Our approach for analysis of the stable isotope data was to assume that changes in concentration of nitrate and ammonium in vertical profiles in the regions of overlap between plumes are due to 1) dilution at edges of plumes, producing no isotopic fractionation, and 2) biogeochemical transformation (loss) due to an isotopically fractionating process. Since each plume is devoid of the other N species, we need not account for mixing of N pools of differing isotopic ratio. Therefore, we used changes in N stable isotopic ratios and in concentration to estimate the proportion of total change in concentration in the vertical profiles that was due to dilution and the proportion due to transformation. Analyzing the profiles in the vertical direction, in terms of mixing of plumes, is analogous to analyzing progress of reactions resulting from progressive mixing that initiated some distance upgradient.

For the region of overlap between nitrate and ammonium plumes in the freshwater aquifer at piezometer 7 (Fig. 6A), we assumed that initial δ^{15}N was equal to the value measured at peak N concentration in the center of the plumes and used isotopic enrichment factors reported in the literature for nitrification (loss of NH_4^+) and denitrification (loss of NO_3^-) (Table 2). In those calculations, we assumed that nitrification and denitrification were one-step processes and ignored any isotopically light nitrate that may have been produced due to nitrification. Decrease in concentration not accounted for by transformations was assumed to be due to dilution of the N plumes at their edges. The result suggests that loss of NO_3^- in the freshwater zone was in the range of 104 μmol L^{-1} (84% of initial concentration), while loss of NH_4^+ was 59 μmol L^{-1} (22% of initial concentration). A similar approach based on NH_4^+ concentration and $\delta^{15}N$ in the deep STZ suggested loss of 18 μmol L^{-1} NH_4^+ (31% of initial concentration) in a location where terrestrial-source nitrate mixed into the saline groundwater zone (Fig. 6D to F; Table 2). The calculations based on the Rayleigh model are not intended as a rigorous measurement of the extent of nitrification and denitrification, but rather as strong indication that DIN transformations occur in the zones examined, and as a rough approximation of magnitude.

It is difficult to translate loss estimates based on the stable isotope measurements to rates of loss or total mass of N lost prior to discharge in part because it is not known over what time interval the changes in isotope ratios occurred. However, we can make those estimates based on change in DIN mass flux (mmol m^{-1} shoreline d^{-1}) in the freshwater zone along the flow-path prior to discharge. Along transect A at piezometer 7, average DIN concentration was 93 μmol L^{-1} in the 6 m thick freshwater aquifer above the deep STZ, while 12 m downgradient and after progressive mixing of the oxidized and reduced plumes, average DIN concentration at piezometers 11 and 3 was 33 μmol L^{-1} in the 3.4 m thick freshwater aquifer (Fig. 2, Web Appendix). Thus, given a discharge rate in this valley portion of the watershed of 1.23 m^3 m^{-1} shoreline d^{-1} (Mulligan and Charette 2006), and assuming conservation of that mass flux through the sampled portion of the aquifer, DIN flux at piezometer 7 was ~ 114 mmol m^{-1} shoreline d^{-1}. Due to decrease in N concentration 12 m downgradient at piezometers 11/3, DIN flux was ~ 41 mmol m^{-1} shoreline d^{-1}. That decrease in mass flux of DIN occurred during ~ 14 d transit between piezometers (calculated based on discharge rate and on aquifer thickness). Thus, assuming that the magnitude of the N source was constant over a 14 d period, we estimate that DIN was lost at a rate of ~74 mmol m^{-1} shoreline d^{-1}, or that 65% of DIN mass flux had been lost. In terms of N concentration, loss rate is estimated at ~ 4.4 μmol L^{-1} d^{-1}. This estimated loss of inventory should reflect losses that appear to have occurred both in the freshwater zone and via mixing of N into in the deep STZ, though it is not possible to determine the relative importance of these two zones to the extent of N removal.

Mechanisms of Nitrogen Transformation—Throughout the seepage zone, the distributions of redox-cycled elements examined in this study are consistent with theoretical thermodynamic stability of chemical species (Stumm and Morgan 1996). For instance, along the salinity gradient in the deep STZ, redox conditions transition from a nitrate stability zone within the immediately adjacent freshwater, to nitrate reduction and ammonium stability zone at low salinity, manganese reduction zone at intermediate salinity, and iron reduction zone at high salinity (Fig. 6, Web Appendix). Similar distributions occur in the freshwater plumes. Infrequent overlap in distribution of nitrate with that of ammonium (and nitrate with Fe^{2+}), suggest that in general the system approaches equilibrium with respect to the nitrate/ammonium redox couple. Such a result suggests that biogeochemical transformation rates are rapid relative to the rate of mixing of dissimilar water masses, a condition which maintains sharp interfaces between biogeochemical zones (Smith et al. 1991, Postma et al. 1991). Therefore, co-occurrences of nitrate with ammonium or Fe^{2+}, which were observed occasionally, are indication of thermodynamically unstable conditions that must be the result of relatively recent mixing processes at interfaces, and changes in natural abundance stable N isotope ratios suggest that where groundwater residence time is sufficient, N transformations do occur.

There has been a proliferation, largely during the past decade or so, of microbially-mediated pathways for oxidation/reduction reactions involving N that have been shown or proposed to occur in aquatic environments (e.g. Luther et al. 1997, Hulth et al. 1999, Schmidt et al. 2002). Microbial processes are the most likely cause for the N loss, as opposed to adsorption to sediment, since the isotopic enrichment factor is positive for adsorption, and would therefore result in a decrease in δ^{15}N of the residual material (Karamanos and Rennie 1978). Given conditions and availability of reactants, mechanisms for apparent concurrent removal of NO_3^- and NH_4^+ in locations of co-occurrence, both in the freshwater aquifer and deep STZ, could be any combination of heterotrophic or autotrophic denitrification, coupled nitrfication/denitrification, anammox, or Mn oxide oxidation of NH_4^+ to NO_3^- or N_2. Denitrification coupled to mineralization of organic matter may be limited in these permeable sediments given low DOC concentration (20 to 60 μmol L^{-1} in the regions of N loss) and low % organic C in sediment (non-detectable to 0.075, Charette et al. 2005), likely coupled with low reactivity of the organic carbon. Denitrification coupled to oxidation of reduced iron may occur (eq. 1; Hulth et al. 1999, Bohlke and Denver 1995), as dissolved and particularly solid phase concentrations of reduced iron are quite high (Testa et al. 2002, Charette et al. 2005), and since decrease in dissolved Fe^{2+} concentration occurs concurrently with nitrate and ammonium loss both in the freshwater and deep STZ (Fig. 6A and 6D).

$$NO_3^- + 5Fe^{2+} + 12H_2O \rightarrow 5Fe(OH)_3 + 1/2N_2 + 9H^+ \qquad (1)$$

Weber et al. (2006) found both Fe oxidizing and Fe reducing bacteria (*Geobacter metallireducens*) in freshwater wetland soils, and demonstrated microbial nitrate reduction by Fe^{2+}, however the product was largely ammonium, rather than N gas. Chaudhuri et al. (2001) did find nitrate reduction to N_2 by Fe^{2+}. Postma (1990) suggests that reduction of nitrate by Fe^{2+}

is catalyzed at the surface of freshly precipitated Fe-oxyhydroxides, which are abundant at the reduced/oxidized boundaries at the Waquoit study site (Charette and Sholkovitz 2002).

Oxic nitrification seems an unlikely explanation for NH_4^+ removal in the region of overlap of the freshwater plumes because in the shallower portion of the NH_4^+ plume at piezometer 7, D.O. concentration is similar to concentration in the zone of NH_4^+ removal (avg. 26 μmol L^{-1} D.O., Fig. 6C), and yet NH_4^+ concentration and $\delta^{15}N$ do not suggest removal in the shallower portion. If nitrification does occur at the edges of the freshwater ammonium plume or low salinity portions of the deep STZ, it must be rapidly coupled to denitrification since there is no evidence for accumulation of nitrate in the zones of NH_4^+ loss. Ammonium oxidation processes can occur under anoxic conditions, coupled to reduction of Mn or nitrite (by the anammox process), leading to production of N_2 gas. Anaerobic ammonium oxidation (anammox) is an autotrophic process originally discovered in a wastewater treatment plant that is carried out by bacteria in the order *Planctomycetales* (Schmidt et al. 2002). In the anammox process, NH_4^+ is oxidized with NO_2^- as the electron acceptor, with the resulting N_2 composed of atoms from both reactants (eq. 2; van de Graaf et al. 1995).

$$NH_4^+ + NO_2^- \rightarrow N_2 + 2H_2O \qquad (2)$$

In the key zones of NO_3^- and NH_4^+ co-occurrence and removal, in-situ energetics calculations suggest that anammox is thermodynamically favored over both oxic nitrification and MnO_2 oxidation of NH_4^+ in this environment (W. Bach, written comm.).

Nitrogen fluxes to Waquoit Bay—To make separate calculations of nutrient fluxes to the estuary due to fresh and saline SGD, we estimated average concentrations of nutrients in the different water masses, and multiplied by estimates of groundwater discharge rate from each water mass. Such a calculation is conceptually simple, but there are complexities involved. First, it is not clear which groundwater samples are most appropriate to include in calculations of average nutrient concentration, or how many samples are required to capture a meaningful average. For instance, groundwater samples collected at several m below the sediment surface, or certainly at an on-shore groundwater monitoring well, may not be relevant to concentrations at the time of discharge, if biogeochemical transformations occur during transit through the intervening meters of sediment, as demonstrated in this study between piezometers 7 and 3. Second, assignment of groundwater and nutrient sources is complicated by the fact that much of SGD along coastal margins commonly occurs as brackish groundwater, including most of the terrestrial-source groundwater and nutrients (Fig. 2; Bokuniewicz et al. 2004). In addition, there is the obvious complexity of separately estimating rates of groundwater discharges from terrestrial and re-circulated marine sources and evaluating associated uncertainty. In the present publication, we do not evaluate complexities related to estimating groundwater discharge rates, but rather accept the estimates (determined for June 2003) from Mulligan and Charette (2006) and Michael et al. (2005) as described in the "Methods" section and refer readers to those publications for detailed discussion of groundwater discharges at this site.

To obtain the most relevant average nutrient concentrations from the available data, and to examine the importance of some of the complexities mentioned, we modified the calculation in two ways: First, in calculating average concentrations we only considered samples collected from within the groundwater discharge zone (intertidal or shallow subtidal) and from < 0.5 m depth below the sediment surface. Of the 328 samples collected for this study, only 15 met those qualifications (Table 3). Second, we classified discharges based on the N sources, terrestrial vs. marine, rather than based on salinity. For the purpose of assigning source to N contained in the shallow groundwater samples, we examined salinity and N concentrations in samples collected nearby and upgradient. As mentioned previously, it is clear at this study site that virtually all N occurring in the shallow STZ is carried into that zone from terrestrial groundwater (Web Appendix, Fig. 2; Talbot et al. 2003). Thus, N carried in the shallow STZ was considered to have a terrestrial source. It is also evident that terrestrial-source N is not carried within the deep STZ, since the freshwater ammonium plume does not intersect the deep STZ (Fig. 2) and since, as discussed earlier, terrestrial nitrate is eliminated at very low salinity within the deep STZ. Thus, N carried within the deep STZ was considered to have a marine (sediment regenerated) source. The result was that 9 shallow-depth samples with an average salinity of 7 were used to estimate nutrient concentration in the terrestrial-source SGD, and 6 samples with average salinity of 26 were used to estimate average N concentration in marine-source SGD (Table 3).

In the Mulligan and Charette (2006) study, freshwater (terrestrial) groundwater discharge rate during summer of 2003 was estimated to be 4 to 7 fold greater than discharge rate of saline groundwater (Table 4). As discussed by the authors of that study, attempts to separately quantify fresh and saline groundwater discharges at other sites have typically found saline groundwater discharge rates to be greater relative to freshwater discharge rates (e.g. Michael et al. 2005, Kroeger et al. 2007). Mulligan and Charette suggested that high rate of fresh discharge, and simultaneous low inventory of radiochemical tracer of saline discharge, during their study might have been related in part to a high water table and unusually large rainfall during the spring and early summer preceding their measurements. The seepage meter study at Waquoit Bay (Michael et al. 2005) resulted in estimated saline discharge rate that was similar to the rate of freshwater discharge (Table 4). Based on the range of estimates of marine-source groundwater discharge in the Waquoit Bay studies, and due to relatively low concentration of DIN in marine-source groundwater, we estimate that advective flux to surface water of regenerated DIN from estuarine sediments due to recirculation of saline groundwater was 4 to 24% of the rate of new DIN loading due to discharge of terrestrial groundwater. We estimate that flux of regenerated PO_4^{3-} due to saline groundwater recirculation was 38 to 224% of the rate of loading due to fresh groundwater discharge. We note, however, that loading of PO_4^{3-} is particularly difficult to estimate, since oxidative precipitation of Fe oxides in the final few cm of groundwater flow path may result in substantial scavenging of PO_4^{3-} from solution prior to discharge (Charette and Sholkovitz 2002).

Conclusions

There is reason to expect that the broad patterns shown here may commonly occur in SGD zones. For instance, mixing of terrestrial-source nitrate into the deep STZ is likely to occur in many locations and to generally lead to nitrate reduction, given the widespread distribution of natural and anthropogenic nitrate in aquifers (Nolan and Stoner 1995), and reducing and NH_4^+ dominated conditions in saline porewater and groundwater at a wide range of sites throughout the world (e.g. Hauxwell et al. 2001, Windom and Niencheski 2003, Addy et al. 2005, Bratton et al. 2004, Burnett et al. 2007, Kroeger et al. 2007). Furthermore, given generally rapid porewater flushing and relatively high wave energy at the sediment surface in shallow STZs (Michael et al. 2005, Robinson et al. 2006), short residence time and low rate of organic matter supply should typically result in limited N transformations in those zones. Exceptions might be in locations where organic matter or other reducing potential is supplied from marsh soils (Addy et al. 2005, Tobias et al. 2001) or detrital material on beaches, or where residence time is longer in the shallow STZ.

Subterranean estuaries might be expected to commonly contain low concentrations of organic carbon, due to mineralization upgradient in the freshwater aquifer, and due to high rates of mineralization maintained by advection through permeable sediments (de Beer et al. 2005) in saline groundwater recharge zones. However, as occurs at Waquoit Bay, deep STZs may typically carry stored reducing potential in the form of ammonium, reduced metals, and in some cases sulfide or methane. Low organic C in combination with high concentrations of DIN and active redox cycling of iron and manganese (Huettel et al. 1998, Charette and Sholkovitz 2002, Windom and Niencheski 2003, Kroeger et al. 2007) in permeable sediments within SGD zones should in general favor autotrophic processes over heterotrophic processes. A wide range exists of possible N transformation pathways, including denitrification coupled to oxidation of organic C or of Fe minerals, nitrification, anoxic ammonium oxidation, and anammox. Isotope pairing experiments and analysis of microbial communities are needed to identify the processes occurring.

DIN loss from the freshwater zone at transect A was estimated to be ~65% during just 12 m transit through the seepage zone. However, the critical point of this study is not the estimated magnitude of loss at this study site. Rather, the point is that despite organic C-poor conditions, the near-shore aquifer and subterranean estuary are biogeochemically active zones, where attenuation of land-derived N loads may occur. Further, the extent of loss is likely controlled in part by degree of mixing between dissimilar water masses prior to discharge: had a greater degree of mixing occurred of the freshwater nitrate plume into the deep STZ, N loss would likely have been greater; in locations (such as at transect C in the present study) where mixing does not occur between oxidized and reduced freshwater plumes, loss is less likely to occur in the freshwater zone. Mixing in the seepage zone is related in part to thinning of the freshwater lens and change in flow direction prior to discharge, a feature common to all SGD zones. Mixing is further related to hydrogeological attributes controlling dispersion both within the freshwater aquifer and at the fresh/saline interface.

We have not examined in this study N transformations that might occur during discharge through the last few cm of flowpath, where groundwater discharges through permeable coastal sediments, in some cases enriched with estuarine detrital, algal and microbial material. Nitrogen losses occurring at the land/sea margin, and not related to fringing wetlands, currently are not accounted for in N loading models or measurements, and are in addition to accumulated N losses, typically amounting to ~75% of N loaded to the watershed, in upgradient segments of watersheds including soil, vadose zone, and aquifer (Valiela et al. 1997).

High-resolution sampling techniques like the one described herein are required to capture the sharp geochemical gradients of coastal aquifer systems, and to examine biogeochemical transformations occurring at the critical but narrow interfaces between water masses. Furthermore, for the purpose of estimating N loads due to SGD, the conclusions reached about nutrient source (terrestrial vs. marine) in each sample collected provide a strong argument for such detailed examination of salinity and nutrient distributions in the coastal aquifer. In the absence of context provided by detailed sampling, the source of nutrients in any brackish sample is ambiguous, and investigators are forced to assume that low salinity brackish groundwater carries terrestrial nutrients (at this site, a correct assumption if collected from the shallow STZ; incorrect if collected from the deep STZ), and that higher salinity brackish groundwater carries marine-source nutrients (correct if collected from the deep STZ; incorrect if collected from the shallow STZ). Since our estimate of DIN concentration in terrestrial-source brackish discharge is ~ 3.5 fold greater than that in marine-source groundwater (Table 3), and since discharge rates of marine and terrestrial groundwater commonly differ, careful assignment of sources is important. Furthermore, clear separation of sources is critical since only the terrestrial source represents new N loaded to coastal waters related to human activity on watersheds.

The range of estimates presented here for the rate of saline SGD highlights the relatively high degree of uncertainty in estimating that component of SGD. Fresh groundwater discharge estimates can be compared against an annual watershed water budget, providing reasonable bounds for the rate. Saline SGD, on the other hand, is controlled by less understood hydrological and oceanographic processes, and a wide range of estimates of discharge seems plausible.

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Table 1. Summary statistics for nutrient concentrations and biogeochemical conditions in the three primary aquifer zones. Concentrations of N and PO_4^{3-} in units of μmol L^{-1}. Dissolved oxygen (D.O.) in units of mg L^{-1}, and Eh as mV.

	n		Salinity	D.O.	Eh	pH	NO_3^-	NH_4^+	DIN	PO_4^{3-}	N:P
Freshwater	91	average	0	2.5	367	6.1	49	19	68	2.7	25
		st. dev.	0	2.3	71	0.6	66	48	114	5.7	
Shallow STZ	21	average	7	5.8	379	6.0	93	13	106	0.8	132
		st. dev.	5	2.5	53	0.7	77	34	111	1.5	
Deep STZ	216	average	24	1.4	172	7.4	0.1	35	35	7.1	5.0
		st. dev.	6	1.1	101	0.3	0.6	11	12	4.7	

Table 2. Calculations of nitrogen loss based on changes in natural abundance stable isotope ratios and using the Rayleigh distillation model. Calculations for the freshwater zone are based on location of co-occurrence of nitrate and ammonium at piezometer 7 (Fig. 6A to C); calculations for the deep STZ based on location in piezometer 6 where freshwater nitrate mixed into the deep STZ (Fig. 6D to F).

	Freshwater zone Nitrate	Freshwater zone Ammonium	Deep STZ Ammonium
Initial conc. (µmol L^{-1})	124	264	57
Final conc. (µmol L^{-1})	11	21	9
Initial $\delta^{15}N$ (‰)	6.6	8.0	5.5
Final $\delta^{15}N$ (‰)	32.0	13.3	13.4
Fraction lost*	0.84	0.22	0.31
Loss (µmol L^{-1})	104	59	18
Dilution (µmol L^{-1})**	9	185	30

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*fraction lost = 1- e^((δ final - δ initial)/ε); where ε = enrichment factor

ε = -13.9 for denitrification (Smith et al. 1991)

ε = -21 for nitrification (Kendall and McDonnell 1998)

**Dilution = decrease in concentration not accounted for by isotopically fractionating loss processes.

Table 3. Salinity, dissolved oxygen concentration, and nutrient concentrations in groundwater samples used to calculate nutrient fluxes to the bay due to terrestrial and marine-source SGD. N and PO_4^{3-} in units of μmol L^{-1}. Dissolved oxygen (D.O.) in units of mg L^{-1}.

	Salinity	D.O.	NO_3^-	NH_4^+	DIN	PO_4^{3-}	N:P
Terrestrial	3	5	187	0	187	0	1246
	5	5	186	0	186	0	18605
	0	n.d.	4	0	4	3	2
	0	3	119	0	119	1	175
	15	1	0	147	147	1	121
	17	8	33	3	36	6	6
	3	1	9	1	10	1	15
	20	6	27	1	28	5	6
	1	7	126	0	126	0	374
average	7	4	77	17	94	1.8	51
Marine	29	0	0	23	23	9	2
	28	0	0	26	26	5	5
	20	1	0	22	22	1	21
	21	2	0	33	33	4	8
	28	1	0	24	24	7	3
	29	1	0	36	36	2	18
average	26	0.9	0	27	27	5	5.6

Table 4. Calculations of nutrient fluxes to the bay due to terrestrial and marine-source SGD.

		Terrestrial (Darcy's Law)[1]	Marine (Ra)[1]	Marine (Rn – Darcy)[1]	Marine (seep meters)[2]
Discharge	(m^3 m^{-1} d^{-1})	4.0	0.6	1.0	3.3
NO_3^- flux	(mmol m^{-1} d^{-1})	304	0	0	0
NH_4^+ flux	(mmol m^{-1} d^{-1})	67	15	27	90
DIN flux	(mmol m^{-1} d^{-1})	371	15	27	90
Terr./Mar.	(%)		4	7	24
PO_4^{3-} flux	(mmol m^{-1} d^{-1})	7	3	5	16
Terr./Mar.	(%)		38	67	224

[1] Mulligan and Charette 2006
[2] Michael et al. 2005

Figure legends

Figure 1. Map of the study site and showing locations of sample collections. “WBNERR” and “Boat House” indicate locations of the Waquoit Bay National Estuarine Research Reserve office and boat house.

Figure 2. Contour plots of (A) salinity, (B) nitrate concentration (µmol L^{-1}), and (C) ammonium concentration (µmol L^{-1}) in the subterranean estuary and groundwater seepage face at the head of Waquoit Bay.

Figure 3. Concentrations of nitrate and ammonium in groundwater samples collected from the deep STZ portion of the seepage zone.

Figure 4. Natural abundance N stable isotope ratios of nitrate (A) and ammonium (B) in groundwater samples collected throughout the seepage zone.

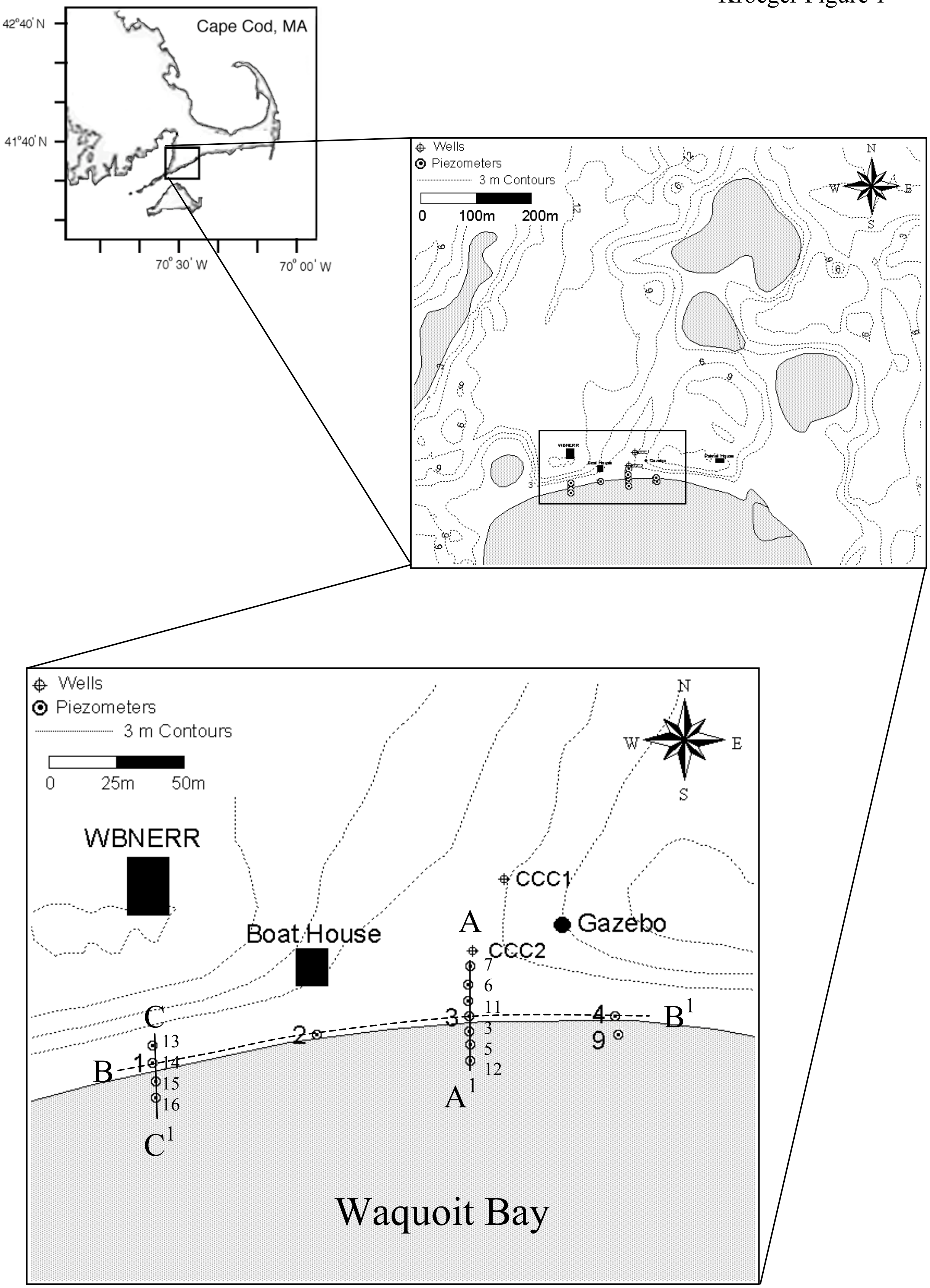
Figure 5. Nitrate concentration vs. ammonium concentration in all groundwater samples collected in this study. Co-occurrence of the two N forms is uncommon.

Figure 6. Depth profiles showing concentration of nitrate, ammonium, dissolved Fe2+; N stable isotope ratios of nitrate and ammonium; salinity and dissolved oxygen concentrations. In transect A at piezometer 7 (A,B,C), overlap in distributions of nitrate and ammonium in the freshwater zone is indicated by the yellow shaded box. In transect A at piezometer 6 (D,E,F)

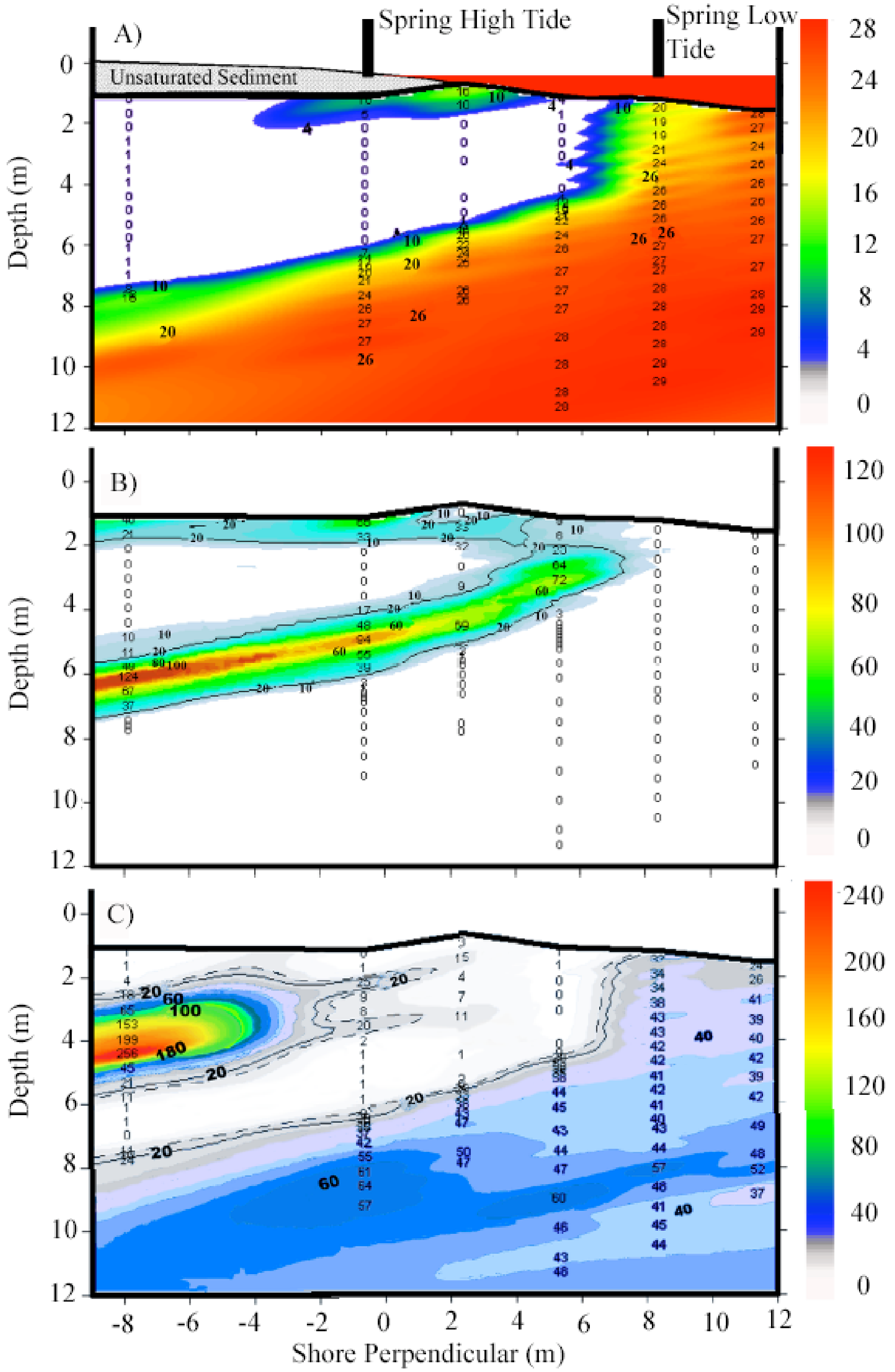
overlap in distribution of nitrate and ammonium in the freshwater zone is indicated by the yellow box at ~3 m depth, and overlap in distributions in the deep STZ is indicated by the yellow box at 5.5 m depth. In transect B at piezometer 4 (G,H,I), overlap in distribution of nitrate and ammonium in the shallow STZ is indicated by the yellow box.

Figure 7. Concentrations and N stable isotope ratios of nitrate and ammonium along the salinity gradient in the deep STZ.

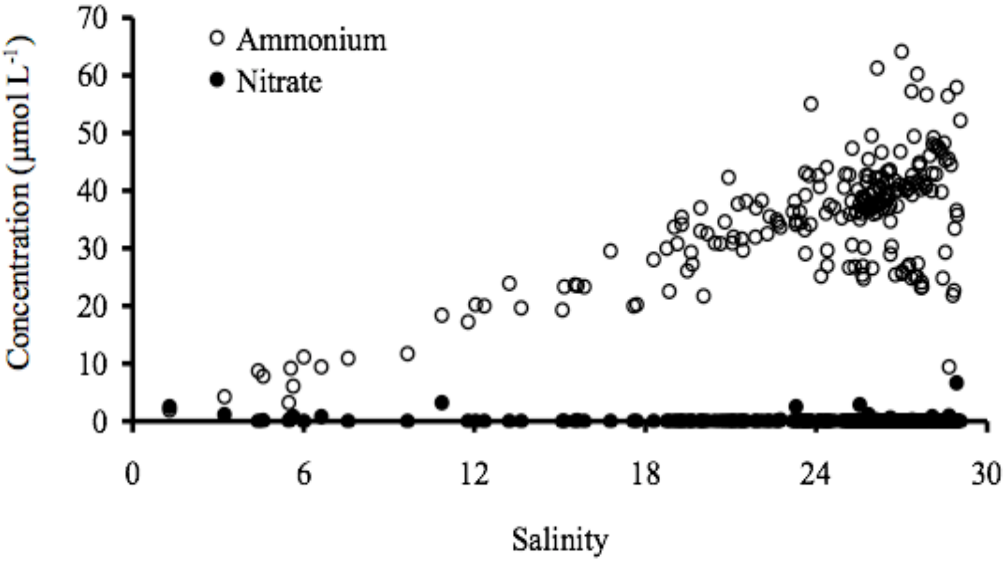
Kroeger Figure 1

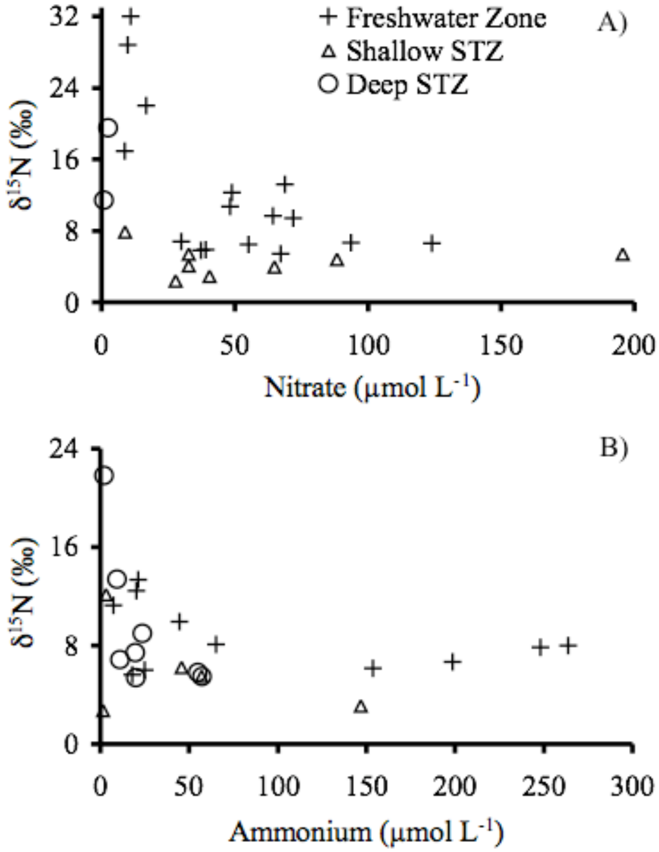


Kroeger Figure 2

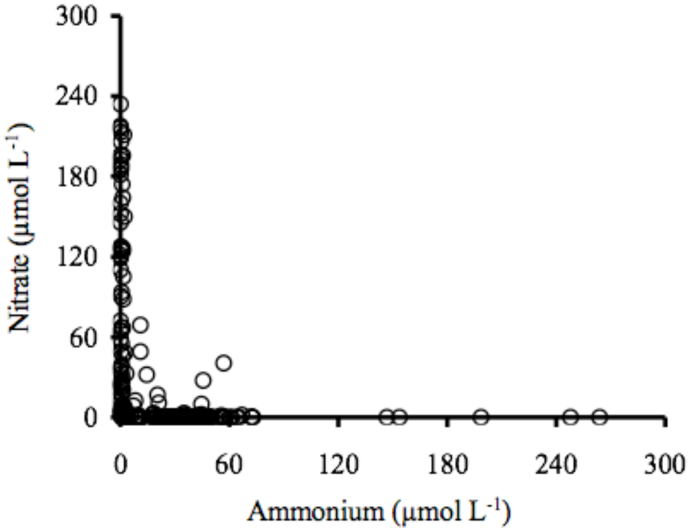


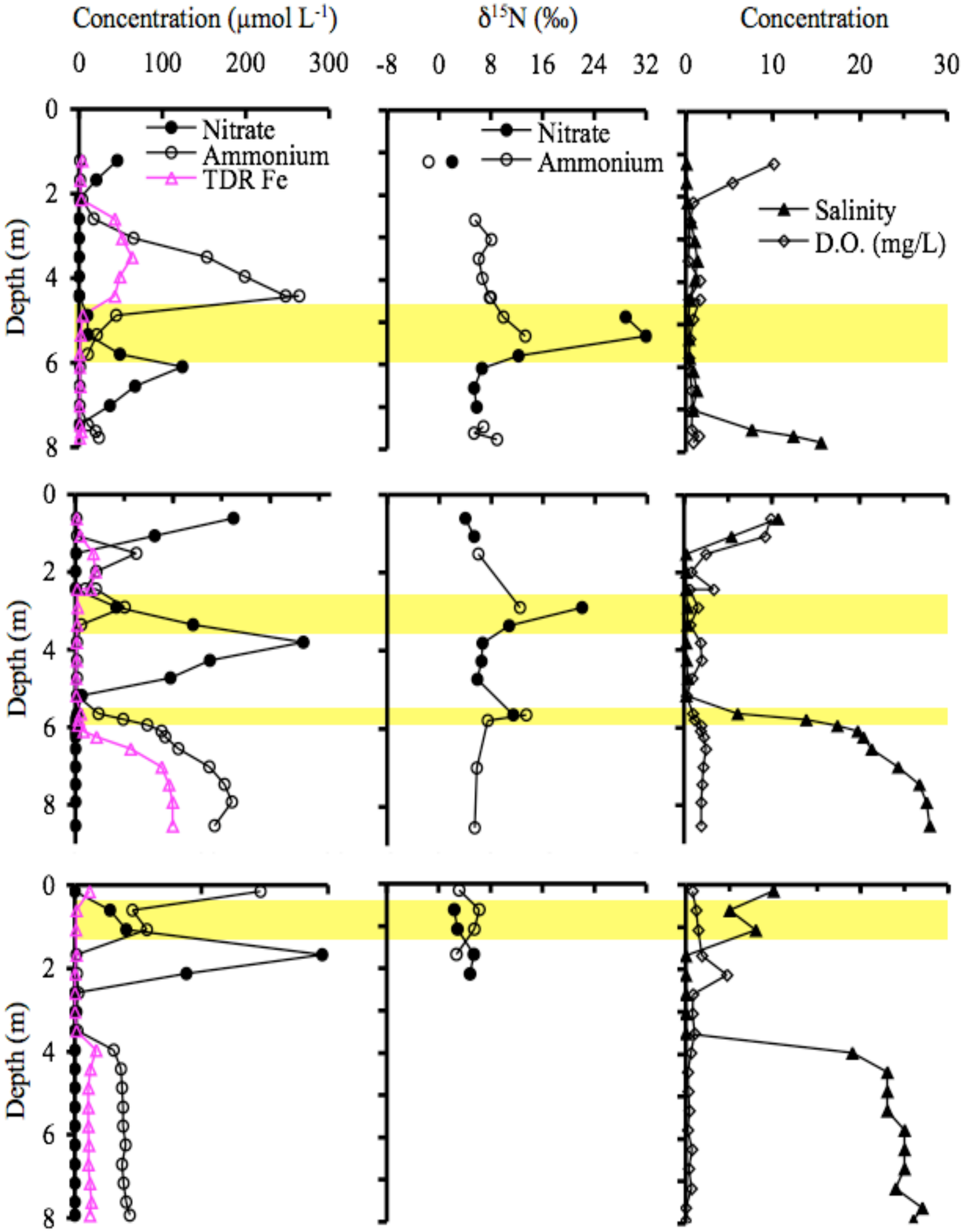
Kroeger Figure 3




32
24
16
8
0
δ15N (‰)
+ Freshwater Zone
△ Shallow STZ
○ Deep STZ
A)
0
50
100
150
200
Nitrate (μmol L-1)
24
16
8
0
δ15N (‰)
B)
0
50
100
150
200
250
300
Ammonium (μmol L-1)

Kroeger Figure 5




Concentration (µmol L-1)
0
100
200
300
δ15N (‰)
-8
0
8
16
24
32
Concentration
0
10
20
30
Depth (m)
0
2
4
6
8
Nitrate
Ammonium
TDR Fe
Nitrate
Ammonium
Salinity
D.O. (mg/L)

Kroeger Figure 7