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University of Maryland Center for Environmental Science

Chesapeake Biological Laboratory

Back River Sediment Flux Measurements to Assess Impacts of Large Reductions in Point Source Nutrient Loading

Final Report

Prepared for: Maryland Department of the Environment

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Executive Summary

We report a synthesis of nutrient loading rates, water-column conditions, and measurements of sediment-water fluxes made in the Back River estuary, Maryland, over the past thirty years. The measurements of loading rates and water-column conditions were made by a number of state and federal agencies, while UMCES scientists measured sediment-water nutrient and oxygen fluxes and sediment nutrient and carbon content. The aim of this study was to examine changes in water-column biogeochemistry, sediment nutrient content, and exchanges of nutrients and oxygen between sediments and the water-column in response to multiple phases of nutrient load reductions from the Back River wastewater treatment facility. We found that total nitrogen loads declined from 7000 kg N day^{-1} in the 1980s to 1500 kg N day^{-1} following the implementation of ENR in 2017. Total phosphorus loads declined by ~90% from peaks in the mid-1980s to the mid-1990s and have been stable ever since. In response, total nitrogen and phosphorus concentrations measured at a sentinel monitoring site since 1985 show declines that generally mirror that of loads from the Back River Wastewater Treatment Plant (WWTP). As a consequence, water-column chlorophyll-*a* concentrations and the frequency of chlorophyll-*a* reaching levels above 100 μg l^{-1} (an intense bloom) have declined modestly over the thirty year period, despite inter-annual variability. This reduction in chlorophyll corresponds to a reduced frequency of nitrogen and phosphorus concentrations that would saturate phytoplankton growth; thus the conditions that might limit phytoplankton growth have become more common. Finally, a collation of sediment biogeochemical measurements made in the mid-1990s, in 2014-2016, and in 2018 following the implementation of ENR reveals that there have been significant declines in sediment-water fluxes of nitrogen, phosphorus, and oxygen and associated sediment concentrations of nitrogen, phosphorus, and carbon. In summary, substantial improvements in wastewater treatment in the Back River estuary led to reduced N and P loads and relatively rapid and substantial reductions in water-column nutrient concentrations, chlorophyll-*a* concentrations, sediment-water nutrient and oxygen fluxes, and sediments stocks of C, N, and P. The recycling of ammonium nitrogen for a given TN load has also declined substantially, consistent with high observed rates of denitrification, indicating that well-mixed estuaries like the Back River recover relatively rapidly in response to nutrient remediation.

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Introduction

Background

Eutrophication, or the enhanced input of organic matter to aquatic ecosystems, remains a pressing ecological problem that is associated with declines in oxygen availability, the loss of submerged macrophyte habitats, and the proliferation of harmful phytoplankton blooms (Kemp et al. 2005). Recognition of this problem has led to expensive and expansive socio-economic commitments to reduce the inputs of bioavailable nutrients that commonly support elevated phytoplankton biomass and associated bottom water and sediment degradation. Although initial efforts to mitigate eutrophication were difficult to associate with clear improvements in tidal waters (e.g., Duarte et al. 2009), sustained nutrient reductions and investments in improved wastewater treatment technologies have led to substantial declines in eutrophication in a growing number of estuaries (e.g., Boynton et al. 2014, Taylor et al. 2011, Stæhr et al. 2017).

While long-term records of water-column variables and their response to eutrophication are common, few long-term records of sediment nutrient content and release to the water column are available (Boynton et al. 2017). Measurements of sediment-water nutrient and oxygen exchanges are particularly important in relatively shallow systems like Chesapeake Bay and its tributaries because the consumption of oxygen (DO) and release of nitrogen (N), phosphorus (P), and silica (Si) compounds at the sediment-water interface have a strong effect on water quality. In much deeper systems (>20 m) the influence of sediment processes is muted and most DO consumption and nutrient re-cycling occurs in the water column (Boynton et al. 2017). Indeed, the very high primary production (mainly via phytoplankton) and secondary production (fish and shellfish) in the Chesapeake is supported, in part, by the efficient sediment re-cycling of essential nutrients (N, P, and Si) supporting primary producers. Upon over-enrichment, however, sediments lose their ability to support secondary production and increase recycling rates of nitrogen and phosphorus to the water-column due to biogeochemical changes resulting from hypoxia and anoxia (Testa and Kemp 2012), further exacerbating eutrophication.

Sediment releases of N and P can be very large in shallow and enriched estuaries such as the Back River, where these nutrient releases serve as basic resources for sustained and excessive phytoplankton biomass and blooms. The Back River watershed and estuary lie immediately north of Baltimore City and the Patapsco River estuary and both areas have been intensely urban and industrial for the past one hundred years (Capper et al. 1983). The Back River sewage treatment facility has been in operation for 104 years, and it historically delivered nitrogen and phosphorus loads that rival the most intensely enriched estuarine systems worldwide. The most obvious sign of eutrophication in this system is the persistent algal blooms that occur at any time of the year (most frequently during summer) when Chlorophyll-*a* concentrations often exceed 100 μg l^{-1}. Improvements in wastewater treatment have led to substantial declines in nutrient loads over time in this estuary, with total phosphorus declining from 700 kg P d^{-1} to 100 kg P d^{-1} between 1984 and 1994 and total nitrogen loads declining from ~7000 kg N d^{-1} to 3000 kg N d^{-1} between 1984 and 1999. A complete assessment of the consequences of these load declines on sediment properties, sediment-water nutrient and oxygen flux, and water-column eutrophication proxies has yet to be accomplished.

The purpose of this study was to make measurements of net sediment-water exchanges of key elements, including oxygen, nitrogen, and phosphorus along the main axis of the Back River estuary following a recent upgrade to the Back River wastewater treatment plant in 2017. We then collated these recent measurements with previous data from the mid-1990s, 2014, and 2015 to assess long-term changes in sediment-water fluxes and sediment-nutrient content. We also collated model estimates of nutrient input from the watershed, wastewater treatment plant (WWTP) nutrient loads, and water-column nutrient and chlorophyll-*a* data collected by the Maryland Department of the Environment, the Department of Natural Resources, and the Chesapeake Bay Program. This evaluation of sediment nutrient releases, as well as the loadings of pollutants from the land and atmosphere, provides information necessary to diagnose the current and future water quality conditions of an estuary, an exercise that typically involves developing, testing and using, in a forecasting mode, various water quality models.

Description of Project and Strategy for Station Locations

We collated available monitoring data from multiple state and federal agencies to characterize nutrient inputs rates and water-column nutrient and chlorophyll-*a* concentrations. These data were generally collected or generated at a monthly timescale. In this report, we describe these datasets in brief, but complete documentation is available for these data from each agency.

For the sediment measurements made in August 2018, the basic sampling strategy was to focus measurements during the summer period when respiration rates and associated nutrient and oxygen fluxes from sediments were very likely to be at or near annual maxima. Measurements of net sediment-water exchanges of nutrients and oxygen, characterization of the nutrient content of surface sediments, water column respiration, and measurement of water quality conditions in near-bottom water were made on a single 2 day sampling cruise to the Back River during 14-15 August, 2018.

Stations were aligned along the main axis of the estuary and along depth gradients and with one station near the mouth of the Back River. We made sediment flux measurements (duplicate cores) at 8 sites in the Back River. We also conducted "blank" measurements at all sites (blanks are core measurements with no sediment used to correct sediment fluxes for water column influences). These water column blank corrections are generally very small. The 8 sites selected is the maximum number of sites that could be measured in 2 days of effort. Given the cost of staff, equipment preparation and boat charges it made sense to do as much as possible while at the site.

All stations were selected to represent the most common water depth in the vicinity of the station. We avoided locating stations in either extremely deep or shallow sites. The goal was to develop a data set that represented as large a spatial area as possible of the Back River estuary.

Acquisition and Analysis of Sediment-Water Oxygen and Nutrient Exchanges

Location of Back River Stations

We visited eight stations in Maryland's Back River estuary during August, 2018 (Fig. 1, Table 1). At all eight stations, measurements of sediment-water oxygen and nutrient exchanges were made along with associated surface and bottom water conditions, surficial sediment chlorophyll-*a*, particulate carbon, nitrogen and phosphorus concentrations, and estimates of bottom water column respiration. We compiled long-term water column chlorophyll-*a* and nutrient concentrations from the surface and bottom water at station WT4.1, which has been routinely monitored at a monthly time scale by the Chesapeake Bay Program (CBP) and Department of Natural Resources since 1984 (Fig. 1). Finally, we analyzed weekly-scale water-column measurements made during 2014-2018 by the Maryland Department of the Environment (MDE).

Sampling Frequency

The sampling frequency was based on the seasonal patterns of sediment water exchanges observed in previous studies conducted in the Chesapeake Bay region (Kemp and Boynton, 1980, 1981; Boynton et al., 1982; Boynton and Kemp, 1985). Consideration was also given to a sampling scheme that was a compromise in terms of cost versus number of measurements. Based on these results the monitoring design adopted for this Back River study involved a single two day sampling cruise to the Back River during 14-15 August, 2018, the time of maximum or near-maximum sediment biogeochemical activity.

Field Methods for Back River Study

Water Column Profiles

At each Back River station, temperature, salinity, dissolved oxygen, turbidity, and fluorescence were measured at surface (0.5 m below the surface) and bottom (approximately 0.5 m above the bottom) using a Yellow Springs Instrument (YSI) EXO2 multiparameter sonde. Turbidity of surface waters was measured using a Secchi disk and light profiles were taken with LI-COR air and underwater quantum sensors to compute light attenuation. Similar instruments (i.e., YSI sonde and sensor package) were used to measure temperature, salinity, and dissolved oxygen in the 1984-2018 CBP and 2014-2018 MDE datasets.

Water Column Nutrients, Chlorophyll-a, and Total Suspended Solids

Near-bottom water samples (~0.5 m above the sediment surface) and surface water samples (0.5 m below the surface) were collected using a high volume submersible pump system. Samples were filtered, where appropriate, using 0.7 μm GF/F filter pads, and immediately frozen. Samples were analyzed by Nutrient Analytical Services Laboratory (NASL at CBL) for the following dissolved nutrients: ammonium (NH_4), nitrite (NO_2), nitrite plus nitrate ($NO_2 + NO_3$), dissolved inorganic phosphorus corrected for salinity (DIP or PO_4) and silica (Si). The 2014-2018 MDE datasets for these variables followed similar protocols and were analyzes at NASL, while the 1984-2018 CBP data set includes some changes in time for analytical techniques and laboratories. Full documentation of the data history can be found on the CBP website (https://www.chesapeakebay.net/what/downloads/cbp_water_quality_database_1984_present).

Sediment Profiles

At each Back River station an intact sediment core was used to measure the surficial (surface to 1 cm depth) sediments by sampling for total and active chlorophyll-*a*, particulate carbon (PC), nitrogen (PN) and phosphorus (PP). This approach was used form the 2018 campaign, as well as the cruises in the mid-1990s, 2014, and 2015.

Sediment Flux Measurements

The Sediment, Oxygen and Nutrient Exchange (SONE) measurement program supported by a variety of environmental agencies has, during the past several decades, made these types of measurements in support of water quality modeling activities. Measurements have now been made in all major (and some minor) tributaries of the Maryland portion of Chesapeake Bay. During 14-15 August, 2018 a single 2 day sampling cruise to the Back River was completed during the time of high sediment biogeochemical activity. All SONE data collected in the Bay region since the late 1970s have been compiled as a large and accessible data set (http://www.gonzo.cbl.umces.edu/). We will add the Back River data collected during 2018 to this large data set in the near future.

The protocol used in making Back River sediment flux estimates included using two replicate sediment cores collected at all stations along with "blank" measurement at all sites. An intact sediment core constituted a benthic microcosm where changes in oxygen, nutrient and other compound concentrations were determined during a fixed incubation time period (usually 3 hours). "Blanks" are core measurements with no sediment used to correct sediment fluxes for water column influences, which are generally very small but occasionally need to be checked.

Intact sediment cores were collected at each station using a modified Bouma box corer when working off the R/V Rachel Carson platform or a modified pole corer when using the R/V Aries. These cores were then transferred to a Plexiglass cylinder (6.5 cm inner diameter x 30 cm length) and inspected for disturbances from large macrofauna or cracks in the sediment surface. If the sample was satisfactory, the core was fitted with an O-ring sealed top containing various sampling ports, and a gasket sealed bottom. The core was then placed in a darkened, temperature controlled holding tank aboard the R/V Rachel Carson where overlying water in the core was slowly replaced by fresh bottom water ensuring that water quality conditions in the core closely approximated *in-situ* conditions.

During the period in which the flux measurements were taken, the cores were placed in a darkened temperature controlled bath to maintain ambient temperature conditions. Cores were arranged around a motorized magnetic "turn-table". The overlying water in the cores was gently circulated with no induction of sediment resuspension via magnetic stir bars attached to the core tops. At some stations we collected a "DO reference core". These cores were fitted with WTW Stir OxG oxygen sensors to monitor the rate of oxygen uptake within the cores. Oxygen concentrations were recorded and overlying water samples (~40 ml) were extracted from each core every 60 minutes during the incubation period. Cores were incubated for 3 hours with a total of 4 measurements collected. As a water sample was extracted from a core, an equal amount of ambient bottom water was added to replace the lost volume. The cores filled with only bottom water (no sediment-"blanks") were also incubated and sampled as described above. Water

samples were filtered and immediately frozen for later analysis for ammonium (NH_4), nitrite (NO_2), nitrite plus nitrate ($NO_2 + NO_3$), dissolved inorganic phosphorous (DIP or PO_4) and silica (Si). Oxygen and nitrogen (N_2) samples were collected in 12 ml exetainers, allowed to overflow, spiked with 10 µl of 30 g L^{-1} mercuric chloride ($HgCl_2$), capped, and stored underwater at incubation temperature. Oxygen and nutrient fluxes were estimated by calculating the rate of change in concentration during the incubation period and converting the volumetric rate to a flux using the volume to area ratio of each core.

Water Column Respiration
Water column respiration measurements were made using a modified biological oxygen demand (BOD) protocol. Whole water samples were taken from the bottom layer at approximately 1 m above the sediment surface using a high volume submersible pump system. Glass dark BOD bottles (300 ml) were gently filled with sample water and allowed to overfill, exchanging the volume at least three times. Triplicate samples were taken for initial and final measurements. Initial samples were read immediately with a ProOBOD® self-stirring optical BOD probe for determination of dissolved oxygen. The same dark BOD bottles were then immediately capped and incubated in a dark ambient flowing seawater incubator. Final samples were read with a ProOBOD® self-stirring optical BOD probe for determination of dissolved oxygen at the termination of the incubation period (~4 hours).

Chemical Analyses used in Back River Study
Methods for the determination of dissolved and particulate nutrients were: ammonium (NH_4), nitrite (NO_2), nitrite plus nitrate ($NO_2 + NO_3$), dissolved inorganic phosphorus (DIP or PO_4), and silica (Si) were measured using the automated method of EPA (1979); particulate carbon (PC) and particulate nitrogen (PN) samples were analyzed using an Elemental Analyzer; particulate phosphorus (PP) concentrations were obtained by acid digestion of muffled-dry samples (Aspila et al., 1976); Total Suspended Solids (TSS) concentrations were obtained by a gravimetric measurement of dry samples; and methods of Strickland and Parsons (1972) and Parsons et al. (1984) were followed for chlorophyll-*a* analysis. Complete analytical methods are included in Table 2. Dissolved gas components, nitrogen (N_2) and oxygen (O_2), were analyzed using the N_2:Ar technique with a membrane inlet mass spectrometer (MIMS) (Kana et al., 1994).

Data Management
QA/QC Field Checks
Cruises and experiments were scheduled well ahead of time with Research Fleet Operations (RFO). A schedule of activities for each day of field work (cruise and experimental plan) was submitted to the PI and other members of the staff. Cruises and experiments that were postponed due to contract negotiations, weather or mechanical problems were rescheduled.

Preparation and Collection of Gear
Prior to initiating a research cruise an experimental run was conducted using all the necessary equipment involved in the collection of water and sediment samples, incubation of sediment cores and collection of physical water quality data and all these items were inventoried using prepared checklists. All equipment was cleaned, checked, and calibrated to insure that it was

fully operational. The equipment was packed into containers for easy transport and loaded aboard vehicles and the research vessels (R/V Rachel Carson and R/V Aries). The checklists were re-examined to verify the presence of all necessary gear.

Standards and reagents involved in the calibration of instrumentation and for chemical analyses in the field were made according to a schedule of shelf life (i.e. daily, weekly or seasonally). All chemicals were handled, prepared and stored in accordance with standard laboratory practices. Safety Data Sheets (SDS) for all chemicals and reagents used were available at all times.

Potential Contamination
During the course of a research cruise different steps were taken to insure that the chances for contamination were minimized. These practices involved almost constant washing of equipment during a cruise. All containers used to collect bulk raw water were rinsed with copious amounts of sample (station) water before they were filled and were thoroughly cleaned with fresh water and dried at the end of the cruise. Containers from which samples were taken for chemical analysis were rinsed additionally with deionized water. The apparatus for taking the sediment samples, as well as the incubation equipment, was thoroughly washed with station water before it was used to collect samples. Upon completion of the cruise the apparatus was rinsed with fresh water. Single use/disposable plastic vials and centrifuge tubes that require no cleaning were used to collect water and sediment samples (after being fully processed) for chemical analysis. All syringes and other laboratory equipment used in processing these samples were washed with deionized water between each use. All glassware associated with the preparation of standards and reagents was cleaned with copious amounts deionized water and acid washed (10% HCL) when appropriate.

Calibration Procedures and Frequency
All instruments involved in the collection of water quality data (temperature, conductivity, salinity, pH, turbidity, fluorescence, and dissolved oxygen) were calibrated daily. Dissolved oxygen calibration incorporates a standard air calibration based on air temperature and barometric pressure. Conductivity/salinity was calibrated with a 0.10 molar standard of potassium chloride. pH was calibrated with buffer 7 and buffer 10. Temperature is calibrated by the manufacturer only when the instrument is returned for service. Turbidity is calibrated with deionized water and a 124 FNU standard. Fluorescence is calibrated with deionized water. All instruments were maintained in accordance with manufacturer's specifications. If any apparent problems arose the instrument was removed from use until the malfunction was diagnosed and remedied. Additional instruments, sensors, and calibration solutions were available at all times in the event of failures.

Recording of Field Data
All field data were recorded on specially prepared field data sheets. The raw data sheets were reviewed for possible missing data values due to sample collection problems. These sheets were scanned and saved, then filed electronically and in the laboratory. A cruise log book was also kept.

Data Tables QA/QC

Data recorded by instruments in the field were entered directly onto specially prepared data sheets. Data from samples analyzed by Nutrient Analytical Services Laboratory (NASL) were returned in electronic format. Data were keyed into the most recent version of Microsoft® Excel. A standard data file format was used. Hard copies of the files were manually checked for errors. Data files were corrected.

Analytical Methods QA/QC Control

The Nutrient Analytical Services Laboratory (NASL) at the Chesapeake Biological Laboratory provides nutrient analyses to University, State and Federal agencies. As part of the laboratory's QA/QC program, NASL participates in cross calibration exercises with other institutions and agencies whenever possible. Refer to http://nasl.cbl.umces.edu and D'Elia et al. (1997) for specific details and updated references but some examples include:

- Particulate carbon and nitrogen cross calibration with Woods Hole Oceanographic Institution and Horn Point Environmental Laboratory.
- International Council for the Exploration of the Sea (ICES) inorganic nutrient round-robin communication. The fourth international inter-comparison report was published in 1991 (Kirkwood et al., 1991).
- Comparisons of dissolved nutrient analyses conducted at Horn Point Environmental Laboratory, Bigelow Laboratory, the University of Delaware and the University of New Hampshire.
- Quarterly cross calibration exercises with Virginia Institute of Marine Science (VIMS) and Old Dominion University (ODU).
- Environmental Protection Agency (EPA) unknown audits for various nutrients have been conducted.
- EPA audits of known nutrients were analyzed using samples in different salinity water while looking for possible matrix effects.

NASL has analyzed National Institute of Standards and Technology (NIST) and National Research Board of Canada reference materials, primarily estuarine sediment, as a check for their particulate and sediment carbon, nitrogen and phosphorus methods.

As part of the Chesapeake Bay Mainstem Monitoring Program, the laboratory routinely analyzes approximately ten percent of the total sample load for QA/QC checks. These samples include laboratory duplicates and spike analyses.

Specific procedures included inorganic nitrogen (ammonium [NH_4], nitrite [NO_2], nitrite plus nitrate [$NO_2 + NO_3$]) and dissolved inorganic phosphorus [DIP or PO_4] for which a standard curve usually comprising five concentrations encompassing the expected range for that particular sample set, were analyzed at the beginning of each new run. A standard, which was treated as a sample, was analyzed at least every 20 samples. Baseline corrections were determined either manually or automatically, depending on the instrument providing the analysis. Data needed to calculate concentrations was recorded along with the sample concentration in laboratory notebooks, a copy of which was provided to our group. This procedure was also carried out for

other parameters performed by the laboratory in support of this effort. Details of precision and limits of detection for the variables are given at https://www.umces.edu/nutrient-analytical-services-laboratory.

Sample Custody

Upon arrival at NASL, samples were counted, observed for potential problems (melting, broken containers, etc.) and placed in a freezer until analysis. Sample information and date of arrival were recorded on a log sheet.

Instrument Maintenance

Analytical instruments are maintained on a regular basis and records are kept of hours of operation, scheduled maintenance, pump tube changes, etc. A critical spare parts inventory is maintained for each instrument. Instrument down-time is minimized by troubleshooting instrument problems telephonically with manufacturers and service representatives. Spare parts can be received within 24 hours via next-day air service.

Calculations

Flux Variables

Core Water Depth (m) represents height of water above the sediment surface in the sediment core chamber.

Core H_2O Depth = (CORE VOL^a/CORE SURFACE AREA^b)/100^c

> Where
>
>> *a*   is the measured volume of water in the sediment core (cm^3)
>>
>> *b*   is the surface area measurement of the core cylinder (cm^2)
>>
>> *c*   converts measurement units to m

General method for calculating net sediment-water fluxes:

*NET DO FLUX (gO_2 m^{-2} d^{-1}) = [(DO SLOPE) * (CORE H_2O DEPTH^a) x (1440^b]*

NET NUTRIENT FLUX (μmoles-N m^{-2} h^{-1}) = [(VARIABLE SLOPE^c) x (Core H_2O DEPTH^a) x (60^d) x (1000^e)]

> Where
>
>> *a*   converts measurements from volumetric to areal basis
>>
>> *b*   converts time units from per minute to per day and from mg to g
>>
>> *c*   variables are NH_4, NO_2, $NO_2 + NO_3$, DIP and Si
>>
>> *d*   converts time units from minutes to hours
>>
>> *e*   converts concentration to moles

Specific calculations:

a. Dissolved oxygen:

$$DO\ FLUX\ (g\ O_2\ m^{-2}\ d^{-1}) = [(DO\ SLOPE) * 1440 * (CORE\ H_2O\ DEPTH)]$$

b. Ammonium:

$$NH_4\ FLUX\ (\mu moles\ N\ m^{-2}\ h^{-1}) = (NH_4^+\ SLOPE * 60 * CORE\ H_2O\ DEPTH) * 1000$$

c. Nitrite:

$$NO_2\ FLUX\ (\mu moles\ N\ m^{-2}\ h^{-1}) = (NO_2^-\ SLOPE * 60 * CORE\ H_2O\ DEPTH) * 1000$$

d. Nitrite plus nitrate:

$$NO_2 + NO_3\ FLUX\ (\mu moles\ N\ m^{-2}\ h^{-1}) = (NO_2 + NO_3\ SLOPE * 60 * CORE\ H_2O\ DEPTH) * 1000$$

e. Dissolved Inorganic Phosphorus:

$$DIP\ FLUX\ (\mu moles\ P\ m^{-2}\ h^{-1}) = (DIP\ SLOPE * 60 * CORE\ H2O\ DEPTH) * 1000$$

f. Denitrification:

$$N_2\text{-}N\ FLUX\ (\mu moles\ N\ m^{-2}\ h^{-1}) = (N_2\text{-}N\ SLOPE * 60 * CORE\ H2O\ DEPTH) * 1000$$

Blank corrections were made by simply subtracting the slope (slope of oxygen or nutrient concentration versus time plot) of the blank core from the slope of the oxygen or nutrient concentration versus time plot for the full sediment core. These corrections were generally either zero or quite small

Criteria for accepting, rejecting and modifying variable slopes used in calculating net sediment water fluxes

Nutrient concentrations were plotted against time of sampling and the slope of this curve is used to calculate net sediment-water exchanges. The following steps assume that all data have been previously verified following normal protocols.

1. If the slope of the nutrient concentrations vs. time plot was statistically significant, the slope was used in calculating fluxes without modification.
2. Occasionally, there are plots which indicated a clear increasing or decreasing trend in concentrations over time but had one observation which diverged strongly (either higher or lower concentration) from the trend. We consider these divergent data to represent contaminated samples (either by addition of the compound or addition of water having a much lower concentration of the compound) and they were not used. The slope was recalculated using lower degrees of freedom and a higher "r" value as a criteria for significance. If the slope is statistically significant, it was used in calculating fluxes.
3. If the concentration vs. time plots were erratic (i.e. no statistically significant increasing or decreasing trend in concentration over time) and if the difference in concentration among variables was greater than twice the detection limit for that variable, the data for

that variable were considered to be non-interpretable. The slope was not calculated and the entry for that variable in the data file was recorded as "NI".

4. If the concentration vs. time plots were erratic (i.e. no statistically significant increasing or decreasing trend in concentration over time) and if the difference in concentration among variables was less than twice the detection limit for that variable, then the slope was taken to be zero and the net sediment-water flux was reported as zero. Occasionally, statistically significant slopes have been found for some variables (mostly nitrite and dissolved inorganic phosphorus) where concentration differences over the incubation period do not exceed twice the reported detection limit. These slopes were used to calculate net sediment-water exchanges.

Results

Strategy for Station Locations

Previous studies in the Chesapeake Bay region have found that strong physical, chemical and biological gradients regularly develop in these systems, even in sub-estuaries as small as the Back River. Because of this, it was prudent to establish a series of sampling stations along the main axis of the estuary and to consider depth gradients as well when selecting sampling locations (Fig. 1).

During the course of our studies in the Back River, beginning in 1994 and concluding in 2018, a total of 13 stations were occupied at various times along the main axis of the estuary in an effort to capture the above indicated gradients and, ultimately, be better able to characterize the system condition as well as responses to management actions. At all 13 stations sediment-water oxygen and nutrient flux measurements were conducted for at least one of the multiple years such measurements were conducted (1994, 1995, 1997, 2014, 2015, and 2018). In addition, water column and sediment physical and chemical characteristics were also sampled whenever sediment-water flux measurements were conducted.

Point Source Nutrient Loads: The Time Series 1985-2018

Annual point source nitrogen (NH_4, NO_2+NO_3 and Total Nitrogen (TN)) and phosphorus (PO_4 and Total Phosphorus (TP)) loads from the Back River Waste Water Treatment Plant (WWTP) are shown in Figure 2.

Clear declines in TN loads were evident even before Biological Nutrient Removal (BNR) began about 1997 and TN loads declined further after BNR was instituted with the exception of a multi-year increase in TN loads between 2002 and 2007 (Fig. 2). In brief, TN loads prior to BNR were between 5000 and 7000 kg N day^{-1}, after BNR between 3000 and 5000 kg N day^{-1}. TN loads were further reduced beginning in August 2017 when the WWTP upgrade to Enhanced Nutrient Removal (ENR) status was completed and are now about 1500 kg N day^{-1}. The combined effects of both BNR and ENR technology have resulted in an almost 5 fold reduction in point source TN loads to this estuary (Fig. 2). The temporary increase in TN loads between 2002 and 2006 is currently unexplained.

In addition to the very substantial TN load reductions there have been substantial changes in the dominant forms of nitrogen released from the WWTP (Figure 2). During the early period (pre-BNR) the dominant nitrogen form released was NH_4, but between 1985 and the initiation of BNR in the mid-1990s, it appears that large amounts of NH_4 were oxidized to NO_2+NO_3 prior to discharge. This resulted in decreasing NH_4 loads and increasing NO_2+NO_3 loads, but stable total inorganic nitrogen ($NH_4 + NO_2 + NO_3$) loads. This pattern continued after BNR was initiated where the main form of discharged nitrogen was as NO_2+NO_3 and when ENR was initiated, discharges of NO_2+NO_3 decreased even further.

The decline in point source phosphorus loads from the WWTP began immediately after 1984 and reached low levels, and remained at low levels from 1994 to the present time (Fig. 2). Total phosphorus load reduction between 1984 and 2018 was approximately a factor of 14, representing a huge decline in this nutrient load component.

Sentinel Station Nutrient and Chlorophyll-*a* Concentrations: Station WT4.1 Results
Various forms of nitrogen and phosphorus in surface waters at the long-term sentinel station (WT4.1) are shown for the period 1985 – 2018 in Figure 3. Clear temporal patterns are evident for all three nitrogen compounds. Over the course of this record TN concentrations declined from almost 1000 μM, a value among the highest N concentrations ever measured in the Bay region, to about 100 μM during summer of 2018. Similar, but smaller, declines were also evident for NO_2+NO_3 and NH_4 (Fig. 3), and NH_4 appeared to decline in concert with WWTP NH_4 load declines. Following ENR, NO_2+NO_3 concentrations were even further reduced. These temporal patterns of nitrogen concentration declines are well correlated with nitrogen load reductions at the WWTP.

Concentration declines in phosphorus were also evident at the sentinel site but the magnitude of change was far less because phosphorus load reductions from the WWTP began at the beginning of the data record. Nevertheless, phosphorus concentrations at WT4.1 were lower, especially for PO_4, at the end of the data record. Finally, PO_4 concentrations exhibited the classic shallow estuarine response of concentrations being higher in summer than in the colder seasons of the year (Fig. 3).

One of the primary reasons for nutrient load reductions is to limit the development of algal blooms which can cause low oxygen conditions and adversely affect seagrass communities. Assessing just when nutrient load reductions limit algal growth is a complex and often expensive exercise involving both laboratory and field experiments. However, some useful insights can be developed concerning algal growth limitation by examining in-situ nutrient concentrations relative to the estimated nitrogen and phosphorus concentrations needed to sustain algal growth at half of the maximum algal growth rate. Such concentrations are referred to as the K_s concentrations or the "half-saturation" concentrations. The frequency at which K_s concentrations were reached for both nitrogen and phosphorus are shown in Figure 4. Interestingly, dissolved nitrogen concentrations always exceeded K_s concentrations (estimated as 5 μM from literature sources) prior to substantial nitrogen load reductions from the WWTP. After 1996, 10% to 25% of nitrogen concentration measurements were less than the nitrogen K_s value and after 2010 all years but 2016 exhibited 10% to 25% of nitrogen concentrations less than K_s values (or 90-75%

above K_s). Finally, during 2018, the first full year of ENR-reduced nitrogen loads, only 60% of dissolved nitrogen concentrations were above the K_s value. The case for phosphorus based algal growth rate limitation is even stronger than it is for nitrogen. Even early in the data record (pre-1998) most years exhibited 95% to 75% of PO_4 concentrations measured at station WT4.1 to be greater than phosphorus K_s value (0.23 μM). Later in the data record, only 85% to 30% of PO_4 concentrations were greater than the phosphorus K_s value. In summary, nutrient load reductions from the WWTP were closely associated with both declines in water column nutrient concentrations at the sentinel station and with increased frequency of nitrogen and phosphorus concentrations that were low enough so they could limit algal growth rates.

Chlorophyll-*a* concentrations recorded at the sentinel station (WT4.1) are shown for the period of record in Figure 5a. It should be noted that chlorophyll-*a* concentrations in the Back River are among the highest recorded in any portion of Chesapeake Bay. Even the three lowest summer values recorded at station WT4.1 (20-60 μg l^{-1}) are higher than the highest values recorded in almost all other areas of Chesapeake Bay. Summer chlorophyll-*a* concentrations ranged from about 20 (2003) to over 200 μg l^{-1} (1997) and in most years ranged between 50 and 100 μg l^{-1}. Perhaps the most striking feature of the summer chlorophyll-*a* record is the degree of inter-annual variability, a degree of variability that far exceeded variability associated with nutrient inputs from the WWTP. Clearly, other factors, in addition to nutrient loads and concentrations, played into generating chlorophyll-*a* concentration patterns. For example, the ratio of NH_4 to NO_2+NO_3 declined substantially over the time-series (WWTP NH_4 loads declined rapidly beginning in 1985) and high relative NH_4 concentrations can selectively favor different phytoplankton species (Glibert et al. 2016). However, chlorophyll-*a* concentrations during summers of 2017 and 2018 were the fourth and third, respectively, lowest concentrations in the entire record.

An additional way to examine the data for temporal chlorophyll-*a* patterns is to count the number of samples in each year that exceed a relevant threshold values (Figure 5b). In this case we chose 100 μg l^{-1} as the threshold value. The frequency at which especially high chlorophyll-*a* values were encountered (>100 μg l^{-1}) was greater prior to BNR at the WWTP than in the most recent decade. The record post-ENR record is very short (late summer 2017 and summer 2018) but just less than 10% of measurements during 2018 were in excess of 100 μg l^{-1}.

In summary, time series data indicating decreased nutrient concentrations, increased frequency of nutrient concentrations being less than K_s values for both nitrogen and phosphorus and decreased frequency of occurrence of very high chlorophyll-*a* values all are strong indicators of a retreat from extreme eutrophication in this system, all associated with nutrient load reductions from the WWTP.

Recent Time-Space Nutrient and Chlorophyll-*a* Concentrations
Beginning in August 2014, the Maryland Department of Environment began making monthly water quality measurements at six sites (M1, M2, WT4.1, M3, M4, and M5) along the axis of the Back River that spanned a section of the estuary from upstream of the WWTP to the mouth of the estuary (see Fig. 1). Key water quality variables are presented as time (x-axis) – space (y-

axis) plots in Figure 6. We note here that this type of data, capturing both time and space variability, are rare in the Chesapeake Bay system and of significant value. In these plots station M5 is the most upstream site and M1 is near the mouth of the estuary.

A number of interesting features emerged from these plots. First, and perhaps most importantly, there were very substantial concentration declines in nutrient (both nitrogen and phosphorus) concentrations and chlorophyll-*a* associated with the institution of ENR during August 2017. This pattern of decreased concentration is especially evident for chlorophyll-*a* and NO_2+NO_3 but changes in PO_4 concentrations were small. Concentrations of NH_4 were low at the beginning of this record and did not change a great deal. An important result here is that substantial load reductions produced an almost immediate response in water column nutrient and chlorophyll-*a* concentration. A second feature of note is that the center of the highest nutrient concentrations was just downstream of the WWTP, as expected, and centered on station M4. However, algal utilization of these nutrients was expressed somewhat farther downstream, centered on stations M4 to M2. This spatial shift likely represents the time needed for the algal community to develop in response to elevated nutrient conditions. Finally, despite relatively huge nutrient inputs and resulting very large algal stocks, concentrations of both nutrients and chlorophyll-*a* were reduced at the mouth of the estuary, indicating a huge capacity to process both algal biomass and nutrients.

Sediment-Water Oxygen and Nutrient Fluxes: The 2018 Summer Survey
A single survey of sediment-water oxygen and nutrient fluxes was conducted in the Back River estuary during August 2018. A suite of sediment-water flux measurements, as well as water column and other sediment measurements were made at 8 stations located along the main axis of the estuary (Fig. 7 and Table 3).

Results (means and standard deviations) of all flux measurements are shown as a series of bar graphs in Figure 7. SOC fluxes ranged from 750 to 1750 μmol O_2 m^{-2} hr^{-1} and were highest at stations DPCK and M3 associated with highest concentrations of water column chlorophyll-*a* (Fig. 6). SOC rates were lower both upstream and downstream of the sites with the maximum SOC rates. Sediment NH_4 fluxes also exhibited maximum rates at station DPCK, were somewhat elevated at an adjacent upstream site (station M4) and were much lower at all the downstream sites. Ammonium fluxes ranged between about 30 and 250 μmol N m^{-2} hr^{-1}. Sediment fluxes of PO_4 ranged between zero (M4) and about 30 μmol P m^{-2} hr^{-1} and there was a very distinct spatial pattern. At the upstream, and freshest portion of the estuary, PO_4 fluxes were very low, below levels that are ecologically significant (i.e., not enough P released from sediments to support substantial algal growth). However, at sites in the middle section of the estuary sediment phosphorus fluxes were substantial (~25–30 μmol P m^{-2} hr^{-1}). Sediment phosphorus releases of this magnitude are sufficient to support substantial algal production. Sediment fluxes of nitrite plus nitrate also exhibited a consistent pattern with fluxes directed into sediments in the upper estuary where water column nitrite plus nitrate concentrations were very high while fluxes were directed, at modest rates, from sediments to the water column in the lower estuary indicative of sediment nitrification activity. With the exception of nitrite plus nitrate fluxes at station M1.5, most fluxes were relatively small. Perhaps the most striking aspect of the sediment flux data set

were the high to very high rates of denitrification observed at all stations and particularly at stations in the middle section of the estuary (where SOC and NH_4 fluxes were high). Denitrification rates ranged from about 150 to about 450 µmol N m^{-2} hr^{-1}. To place these high rates in some perspective, earlier nitrogen budgets for the mainstem Chesapeake Bay reported denitrification rates of 40 µmol N m^{-2} hr^{-1}, about an order of magnitude lower than the maximum rates reported here. Rates of this magnitude exert a strong influence on the nitrogen economy of even a very eutrophic ecosystem such as the Back River estuary.

Sediment-Water Oxygen and Nutrient Time-Series: Major System Responses

In a recent global assessment of estuarine and coastal sediment-water oxygen and nutrient fluxes it was reported that time series of such data were extremely rare. In fact only a few systems were found where sediment flux measurements were made for more than two or three years (Boynton et al. 2017). Thus, the six year record for the Back River makes this a special data set and it becomes even far more special because the sediment-water flux time-series encompasses two significant nutrient load reductions at the WWTP. We have summarized oxygen, nitrogen and phosphorus fluxes for the combined mid-1990s period (1994, 1995 and 1997) and for 2014, 2015 and 2018. Further, we have grouped the flux data into upper (stations DPCK, M3.5 and M3) and lower (MDGT, WCPT, WT4.1 and WT4.2) estuarine sites to better relate the flux data to the location of WWTP discharge (Fig. 8). SOC rates in both the upper and lower estuary group declined between the mid-1990s and 2018 and reductions were very large in the lower estuary group. SOC rates in the lower estuary declined by almost a factor of three between the mid-1990s and 2018. As a consequence, SOC only contributed 7-27% of total water column respiration in 2018 (considering measured water-column respiration rates), which is roughly half of the percentage expected for a system this shallow. The pattern of sediment phosphorus fluxes was less clear. However, fluxes were at a minimum during the 2018 sampling at the upper estuary sites and also lower than two of the previous three time periods in the lower estuary. There was a very clear and consistent decline in sediment ammonium fluxes, and of similar magnitude, in both the upper and lower estuary. It is important to note that ammonium fluxes in the mid-1990 period were very large (~ 400 µmol N m^{-2} hr^{-1}) compared to global average rates of about 100 µmol N m^{-2} hr $^{-1}$. Sediment ammonium fluxes in the lower estuary were reduced to ecologically insignificant levels by 2018. For ammonium, SOC, and phosphate, sediment-water fluxes in 2018 were significantly lower than the mid-1990s values ($p<0.05$; Dunn Kruskal-Wallis Multiple Comparison Test). There were two features of nitrite plus nitrate fluxes that were distinctive. First, except for mid-1990 and 2014 fluxes directed into sediments in the upper estuary, all of these fluxes were relatively small. Second, there was a shift, likely related to decreasing NO_3 concentrations in the water column during the period of record, from nitrite plus nitrate fluxes directed into sediments to fluxes directed out of sediments, although at small rates. The sediment to water fluxes likely represent the result of sediment nitrification of interstitial water ammonium. The consistent decreases in sediment-water fluxes, particularly oxygen and ammonium fluxes, are consistent with a system moving from a state of extreme eutrophication to a less, but still very enriched, state.

Sediment C, N, P and Chlorophyll-*a* Concentrations: Is There Sediment Memory?
In these shallow systems the primary organic matter and nutrient storage site is in the sediments. Thus, it is of interest to examine sediment organic matter (POC) and nutrient (PN and PP) concentrations in Back River sediments (Fig. 9 and Table 4). In this analysis we grouped the data into three time periods (mid-1990s, 2014 plus 2015 and the post ENR 2018 year) and further compared sediment concentration data from the Back River with measurements made elsewhere in Chesapeake Bay at sites with low salinity comparable to those in the Back River. Several important features were immediately evident. First, all sediment variables were highest during the pre-BNR period of the mid-1990s. Second, sediment data after the mid-1990s were all lower, likely a response to reduced nutrient loading rates following initiation of ENR at the WWTP. For PC, PN, and PP, concentrations in 2014-2015 and 2018 were significantly lower than the mid-1990s values ($p<0.01$; Dunn Kruskal-Wallis Multiple Comparison Test), as the most recent (2018) sediment data were not lower, or only very slightly lower, than during the 2014-2015 time periods. It seems reasonable to suggest that sufficient time had not elapsed since initiation of ENR at the WWTP to effect sediment organic matter and nutrient concentrations. Finally, for all sediment variables and for all time periods sediment organic matter and nutrient concentrations in the Back River were higher, and in several cases 2 to 4 times higher, than in sediments from , other low salinity sites in Chesapeake Bay. This finding is consistent with the fact that nitrogen and phosphorus loading rates, primarily from the Back River WWTP, have been very high for many decades and that the sediments have accumulated nutrients as a consequence. However, it remains uncertain how chemically and biologically active these sediment storages of particulate organic matter and nutrient are at present.

Discussion and Conclusions

It is clear from our analysis that enhancements to nutrient removal technologies at the Back River WWTP have resulted in measurable reductions in nitrogen loading to this estuarine ecosystem. WWTP load reductions are especially relevant in the Back River estuary, where they represent 89-98% of the total watershed loads of N to the ecosystem (based upon Phase 6 Watershed Model estimates of non-point source inputs). WWTP TN loads have declined roughly 50% following BNR, which is proportional to a 50% decline in TN concentrations in the estuary (~500 to 250 µM) during the same period. In the year since ENR was established in 2017, TN loads have declined by an additional 50%, from a mean of 3157 kg d^{-1} between 2007-2017 to 1608 kg d^{-1} in 2018, coinciding with another sharp decline in TN and NO_2+NO_3 that resulted in NO_2+NO_3 remaining below 50 µM throughout 2018. Clearly, in-estuary properties are tightly connected to WWTP loads and the water column is highly responsive to nutrient load reductions.

The relationship between phosphorus loading and water column concentration was not as clear as for nitrogen, and may be related to sediment-water fluxes. WWTP TP load reductions accomplished in the early 1990s were on the order of 70-80%, but TP and PO_4 concentrations only declined by roughly 50% (Figs. 2 and 3). Similar discrepancies between load reductions and concentrations for phosphorus have been observed in other systems (e.g., Kubo et al. 2019) and these and other authors have cited sustained phosphorus releases from sediments (i.e., internal loading) as a source of P to maintain higher water-column concentrations for several years

following load reductions. In the Back River, surficial sediment P concentrations declined by 50% from the mid-1990s to 2014-2018 and sediment-water fluxes were variable, but moderately reduced over the same time-period, indicating that sustained 'internal loading' for P did not occur. However, the average estuary-wide August sediment-water P fluxes we observed of ~10 μmol P m^{-2} hr^{-1} in 2018 would translate to an estuary-wide input from sediments of 119 kg P d^{-1}, which is roughly equivalent to the current WWTP input rate. Clearly sediment-water phosphorus inputs are an important aspect of the P budget of the Back River.

Reductions in estuarine nutrient concentrations associated with WWTP nutrient input reductions appeared to have reduced the bloom potential for phytoplankton in the Back River. Average summer chlorophyll-*a* concentrations gradually declined over the 1985-2017 period and concentrations were especially low in 2018 (following ENR). This is consistent with an increased tendency for both dissolved N and P concentrations to be at levels potentially limiting phytoplankton growth during summer (Fig.4) and the reduction in bloom frequency observed during the 1985-2017 period. In 2018, following ENR, few chlorophyll-*a* concentrations over 100 μg l^{-1} were measured consistent with the idea that load reductions translated into reduced chlorophyll-*a*. This pattern of WWTP loads leading to modest reductions in chlorophyll-*a* has now been observed in a growing number of estuarine ecosystems (Boynton et al. 2014, Stæhr et al. 2017, Kubo et al. 2019, Riemann et al. 2016), indicating that even in severely enriched systems, eutrophication reversal can occur within a few years of load input declines. Despite often elevated sediment organic matter and particulate nutrient concentrations, "eutrophication memory" seems to be very short in these shallow estuarine systems.

The reductions in sediment nitrogen concentration and flux were much more substantial than for P, consistent with previous modeling and experimental analysis. Sediment nitrogen content declined by 25% from the mid-1990s to 2014-2018 and sediment-water NH$_4$ fluxes declined by nearly 75% in all regions of the estuary. This change could be the result of two possible changes in sediments. First, the declines in sediment PN clearly indicate that less organic nitrogen was available to be remineralized to NH$_4$ (thus reducing the potential flux), but the persistence of relatively high PN content in the sediment suggests that a pool of recalcitrant material has persisted. Another mechanism that could have driven reduced fluxes of NH$_4$ is that nitrification rates in sediment increased. This mechanism is likely in the Back River, where reduced overall inputs of organic matter to sediments (via reduced water-column TN, TP, chl-a) would have led to increased oxygen penetration into sediments and elevated nitrification rates, which would deplete porewater NH$_4$ and lead to reduced sediment-water NH$_4$ fluxes. This pattern would also be consistent with the fact that sediment-water NO$_2$+NO$_3$ fluxes decreased or became slightly positive over time, which could have resulted from elevated NO$_2$+NO$_3$ production in sediments (via nitrification) that led to near-surface porewater NO$_2$+NO$_3$ concentrations becoming closer to or greater than those in the overlying water. Elevated nitrification rates would also generate NO$_2$+NO$_3$ to support coupled nitrification-denitrification, which is likely given the very high rates of apparent denitrification measured in the Back River (150-450 μmol N m^{-2} hr^{-1}).

Cross-system analyses provide an opportunity to place the changes we observed in the Back River into the context of other enriched coastal ecosystems. Across Chesapeake Bay tributaries,

there is a strong positive relationship between annual TN loads and summer NH_4 sediment-water fluxes (r^2 = 0.35, p <0.001; Fig. 10). Back River NH_4 fluxes across all years of measurement are consistent with the Bay-wide pattern, with relatively high TN loads and NH_4 fluxes, but when fluxes and loads are considered on an annual basis, NH_4 fluxes in 2014 and 2015 for a given TN input are far below those measured in the mid-1990s (Fig. 10). This pattern would be consistent with the idea that despite relatively modest TN load declines, local PON production to support remineralization was reduced in recent years, which is consistent with lower chlorophyll-*a* in 2014 and 2015 relative to 1995 and 1997 (Fig. 5) and gradual reductions in sediment oxygen consumption between the 1990s and 2014-2018. Furthermore, the NH_4 fluxes measured for a given TN load in 2018 fell far below the line, consistent with the low chlorophyll-*a* in 2018 and low SOC, revealing a trajectory towards relatively low TN inputs and sediment-water fluxes on a Bay-wide scale.

Net denitrification rates measured in 2018 indicate very high levels and suggest a substantial loss term for nitrogen in the Back River. Rates of net denitrification of 150-450 μmol N m^{-2} hr^{-1} are among the highest reported in the literature for Chesapeake Bay (e.g., Kana et al. 1994, Cornwell et al. 1999) and suggest that the Back River retains a substantial fraction of the nitrogen it receives. Within a global context, denitrification rates are positively related to TN input, which reflects two impacts on elevated N inputs: (a) high overlying water NO_2+NO_3 concentrations to support direct denitrification in underlying sediments, and (b) elevated rates on PON production via phytoplankton that support denitrification (a heterotrophic process) in sediments. Back River denitrification rates for a given TN input are much higher than those in other coastal ecosystems, and in fact, mean denitrification rates at their summer peak levels would be sufficient to remove 100% of the TN load to the Back River from the WWTP, based on 2018 observations (Fig. 11). These high denitrification rates in the Back River arise from well-mixed conditions, which allow for oxygenation and high rates of nitrification, elevated organic matter, and high availability of NO_2+NO_3 that prevent denitrification rates from being limited by nitrification or NO_2+NO_3 availability like anoxic mid-Bay sediments (Testa and Kemp 2012). Although the high summer denitrification rates we measured are probably not representative of the entire year (and thus denitrification probably does not remove 100% of the external input), preliminary model simulations suggest that annually averaged rates approach 225 μmol N m^{-2} hr^{-1}, which is still a substantial rate. The management implications of this finding include the expectation that TMDL-induced increases in oxygen availability should help elevate nitrification to support additional denitrification in sediments.

In conclusion, a collation of measurements spanning three decades indicates that substantial reductions in WWTP N and P loads to the Back River has resulted in relatively rapid and substantial reductions in water-column nutrient concentrations, chlorophyll-*a* concentrations, sediment-water nutrient and oxygen fluxes, and sediments stocks of C, N, and P. The recycling of ammonium nitrogen for a given TN load has declined substantially, consistent with high rates of denitrification, indicating that well-mixed estuaries like the Back River recover relatively rapidly to nutrient remediation. Future work should consider modeling the long-term response of sediments to reductions of PON and POP deposition, develop a river-wide nitrogen and phosphorus budget, and consider the reactivity of the pools of organic N and P that remain in

Back River sediments at high concentrations, but do not appear to significantly affect contemporary sediment-water fluxes.

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Tables

Table 1. 2018 sampling station locations and details.

Station	Depth, m	Latitude	Longitude	Description
M5	1.4	39.298880	-76.482780	Back River at Back River bridge on Eastern Ave(MD rt 150) *Mind cable & pipeline area
M4	1.5	39.295120	-76.474910	Back River Mid-river between Bread & Cheese Ck & Whittaker's Marina
M3	2.5	39.282250	-76.455917	Mid-River off Walnut Pt
WT4.1	1.7-2.5	39.274317	-76.443550	Back River 600 yds ENE Stansbury Pt; 300 yds WSW of R-12
WT4.2	1.6-2.5	39.266300	-76.442267	Upriver of Witchcoat Pt
M2	2.7	39.249400	-76.437417	Back River Mid-river between Porter Pt & Lynch Pt
M1.5	2.3	39.242067	-76.419967	Between M1 & M2; S of Claybank Pt.
DPCK	1.8	39.286217	-76.46205	Mid-River halfway between Cox Pt & Walnut Pt

Table 2. List of water column and sediment nutrient, carbon, and chlorophyll-*a* analysis methods and method detection limits (MDL).

Matrix	Parameter (Units)	Analytical Method	MDL***
Water	Ammonium (NH_4^+, μM)	Standard Methods 4500-NH3 G-1997	0.071 μM
Water	Silica (Si, μM)	EPA 366.0, SM4500-SIO2 C97,11	0.356 μM
Water	Total Suspended Solids (TSS)	EPA 160.2, SM2540 D-1997	2.4 mg l^{-1}
Water	Active Chlorophyll-*a* (μg l^{-1})	EPA 445.0, SM10200H.3	0.69 μg l^{-1}
Water	Total Chlorophyll-*a* (μg l^{-1})	EPA 445.0, SM10200H.3	0.68 μg l^{-1}
Sediment	Active Chlorophyll-*a* (μg l^{-1})	EPA 445.0, SM10200H.3	0.71 μg l^{-1}
Sediment	Total Chlorophyll-*a* (μg l^{-1})	EPA 445.0, SM10200H.3	1.37 μg l^{-1}
Water	Dissolved Inorganic Phosphorus (DIP; μM)	EPA 365.1	0.019 μM
Water	Nitrite (NO_2^-, μM)	EPA 353.2	0.050 μM
Water	Nitrite + Nitrate ($NO_2^- + NO_3^-$ μM)	Reduction via Copper-cadmium or Enzyme Reductases (EPA 353.2)	0.05 or 0.41 μM
Sediment	Sediment Particulate Carbon (%[wt])	EPA 440.0	0.13 %
Sediment	Sediment Particulate Nitrogen (%[wt])	EPA 440.0	0.008 %
Sediment	Sediment Particulate Phosphorus (%[wt])	ASPILA, EPA 365.1	0.009 %

Table 3. Sediment oxygen and nutrient flux results and overlying water (bottom water) concentrations from the 2018 Back River Estuary study.

Station	Date	Time	Station Depth, m	N$_2$-N Flux, µmol N m^{-2} hr^{-1}	N$_2$-N Bottom Water	NH$_4$ Flux, µmol N m^{-2} hr^{-1}	NH$_4$ Bottom Water, µM N	NO$_2$+NO$_3$ Flux, µmol N m^{-2} hr^{-1}	NO$_2$+NO$_3$ Bottom Water, µM N	PO$_4$ Flux, µmol P m^{-2} hr^{-1}	PO$_4$ Bottom Water, µM P	SOC Flux, µmol O2 m^{-2} hr^{-1}	SOC Bottom Water, µM O$_2$
M5	8/14/2018	10:30	1.3	143.54	ND	97.13	10.07	-65.24	41.69	3.30	0.38	-1082.65	176.56
M4	8/14/2018	13:32	1.4	280.24	ND	185.17	1.57	-75.20	30.27	0.07	0.09	-1269.90	195.63
M3	8/14/2018	12:00	2.4	462.09	ND	56.66	0.29	-24.31	0.30	29.31	0.40	-1673.11	213.44
WT4.2	8/14/2018	14:00	2	414.83	ND	29.00	0.36	104.43	0.11	29.41	3.34	-1242.09	250.31
M2	8/14/2018	16:00	2.8	288.50	ND	37.70	0.36	45.75	0.22	11.34	3.34	-1198.65	272.19
DPCK	8/15/2018	7:41	1.5	470.74	ND	263.59	2.00	-33.00	22.13	2.27	0.10	-1812.43	228.13
WT4.1	8/15/2018	9:32	2.2	168.22	ND	35.70	0.57	18.71	0.74	24.56	2.90	-829.99	224.38
M1.5	8/15/2018	7:50	2.8	356.18	ND	51.73	0.29	181.54	4.03	7.05	2.23	-1059.84	217.19

Table 4. Sediment properties measured in the 2018 Back River study.

Station	Date	PN,%	PC, %	PP, %	Active Chl-a, mg m^{-2}
DPCK	8/15/2018	0.49	4.91	0.16	29.33
M1.5	8/15/2018	0.36	3.55	0.12	31.74
M2	8/15/2018	0.46	4.16	0.15	104.92
M3	8/14/2018	0.51	5.06	0.16	37.05
M4	8/14/2018	0.41	3.95	0.18	94.55
M5	8/14/2018	0.28	3.38	0.12	83.21
WT4.1	8/15/2018	0.48	4.48	0.17	44.81
WT4.2	8/14/2018	0.46	4.37	0.15	78.76

Table 5. Water column concentrations measured in the surface water (S) and bottom water (B) in the 2018 Back River study.

Station	Layer	Date	NH$_4$, µM N	PO$_4$, µM P	NO$_2$+NO$_3$ µM N	Active Chl-a, µg l^{-1}	TSS, mg l^{-1}
DPCK	B	8/15/2018	2.00	0.10	22.13	78.82	37.00
DPCK	S	8/15/2018	1.64	0.11	24.34	36.86	32.00
M1.5	B	8/15/2018	0.29	2.23	4.03	28.73	55.00
M1.5	S	8/15/2018	0.29	2.19	0.26	25.35	38.00
M2	B	8/14/2018	0.36	3.34	0.22	57.43	51.00
M2	S	8/14/2018	0.29	3.25	0.34	34.58	53.00
M3	B	8/14/2018	0.29	0.40	0.30	74.61	22.60
M3	S	8/14/2018	0.93	0.15	2.10	51.10	28.00
M4	B	8/14/2018	1.57	0.09	30.27	46.64	49.00
M4	S	8/14/2018	0.43	0.07	33.77	50.95	37.00
M5	B	8/14/2018	10.07	0.38	41.69	52.42	57.00
M5	S	8/14/2018	8.92	0.18	33.70	36.48	56.00
WT4.1	B	8/15/2018	0.57	2.90	0.74	39.57	26.00
WT4.1	S	8/15/2018	0.21	2.88	1.05	55.74	40.00
WT4.2	B	8/14/2018	0.36	3.34	0.11	58.87	36.00
WT4.2	S	8/14/2018	0.86	3.53	0.14	45.21	41.00

Figures
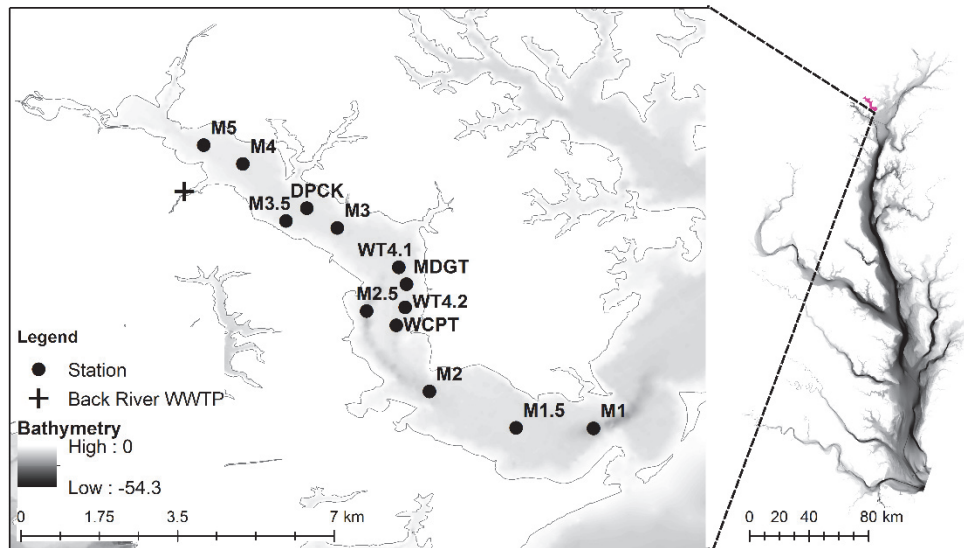
Figure 1. Map of all sampling stations in the Back River estuary, location of the Back River wastewater treatment plant (black cross), and bathymetry in meters for the Back River Estuary and Chesapeake Bay. The location of the Back River within Chesapeake Bay is highlighted in pink. Stations visited in the 2018 study were: M5, M4, DPCK, M3, WT4.1, WT4.2, M2, and M1.5.
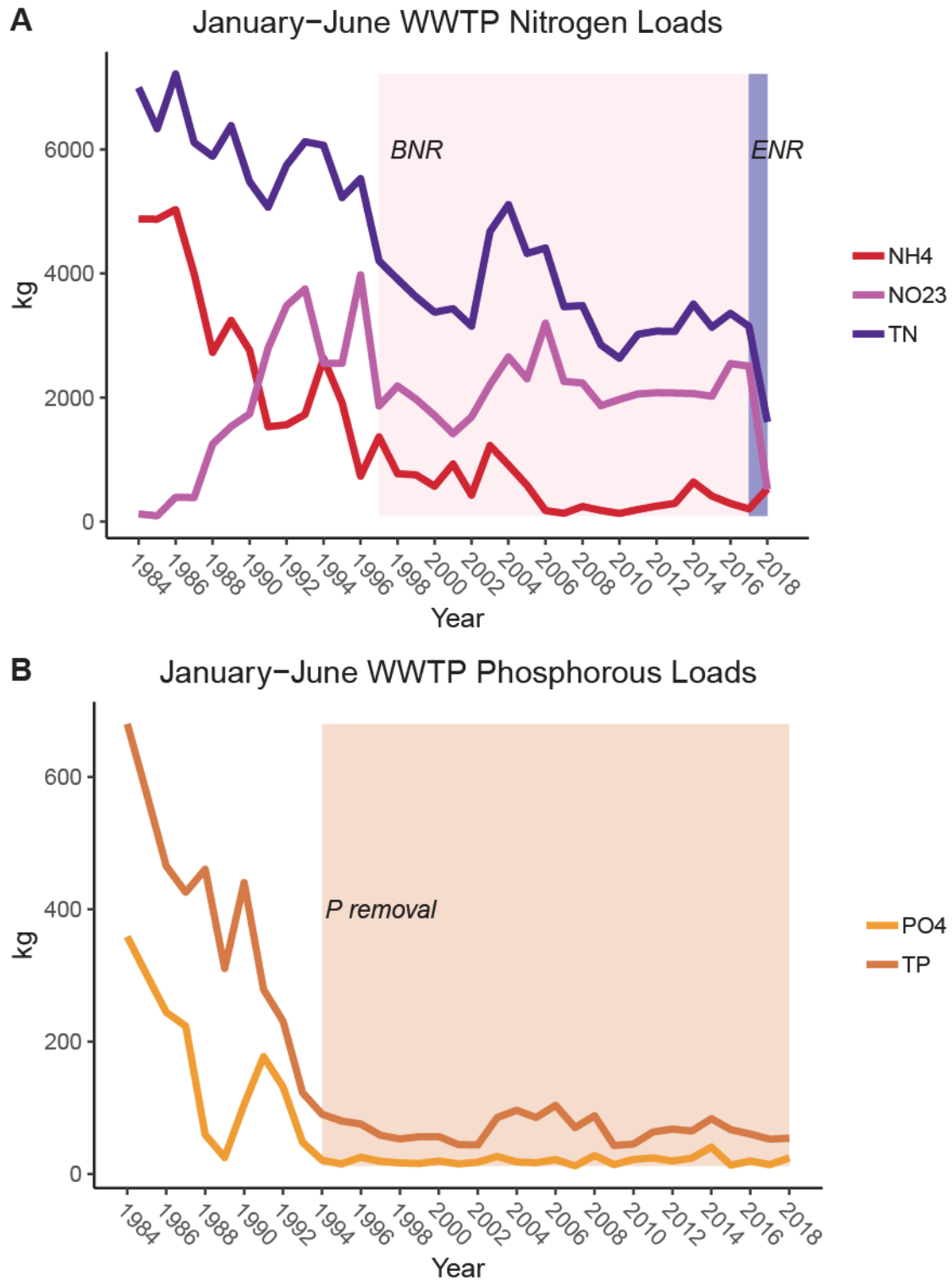
Figure 2. Time-series of average January-June Back River WWTP loads for nitrogen (panel A) and phosphorous (panel B). Shaded areas represent upgrades at the Back River Wastewater Treatment Plant. BNR = Biological Nutrient Removal, ENR = Enhanced Nutrient Removal.
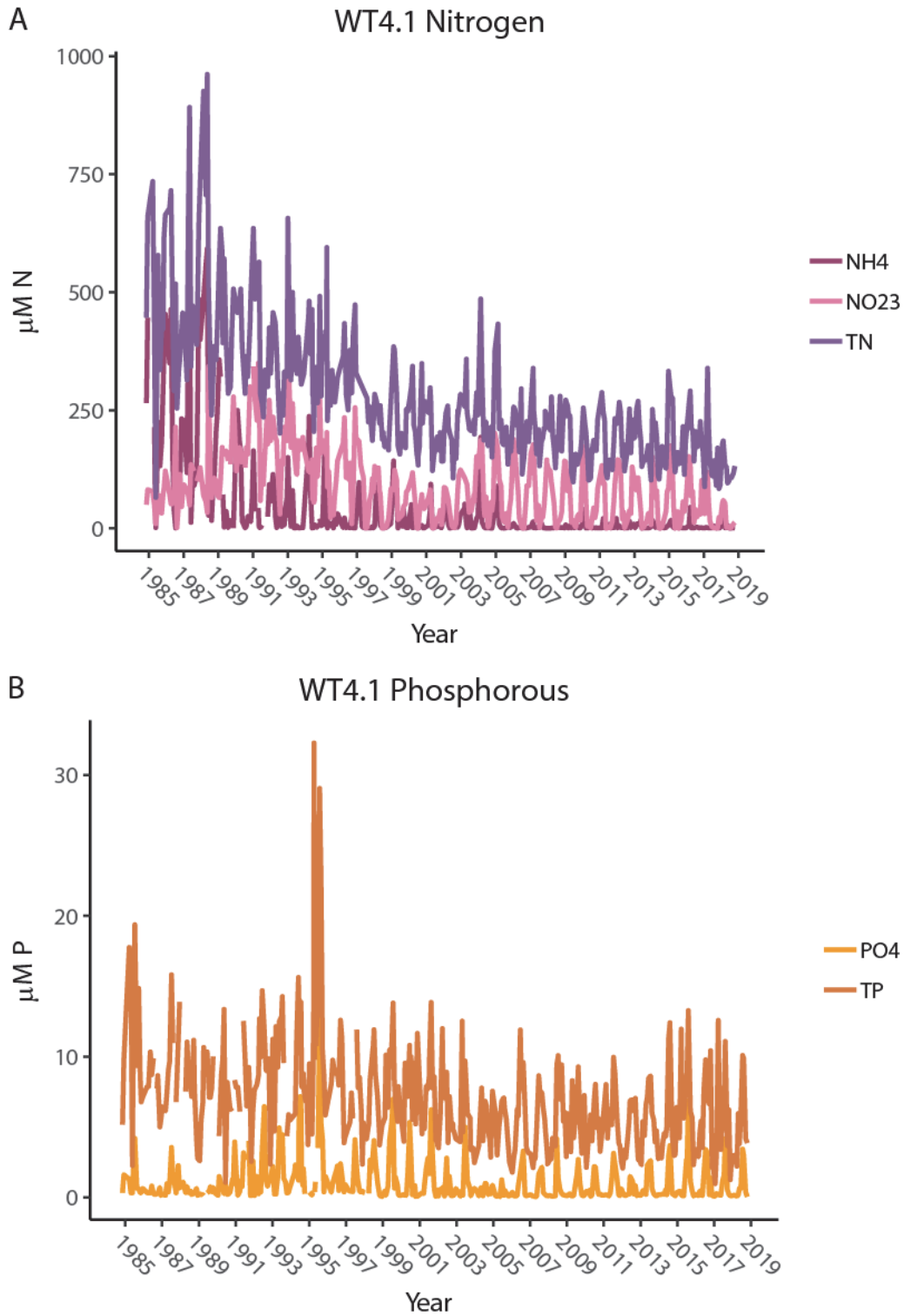
Figure 3. Time-series of monthly total and dissolved nitrogen (panel A) and phosphorous (panel B) concentrations in surface waters at a long-term monitoring station WT4.1 in the Back River.
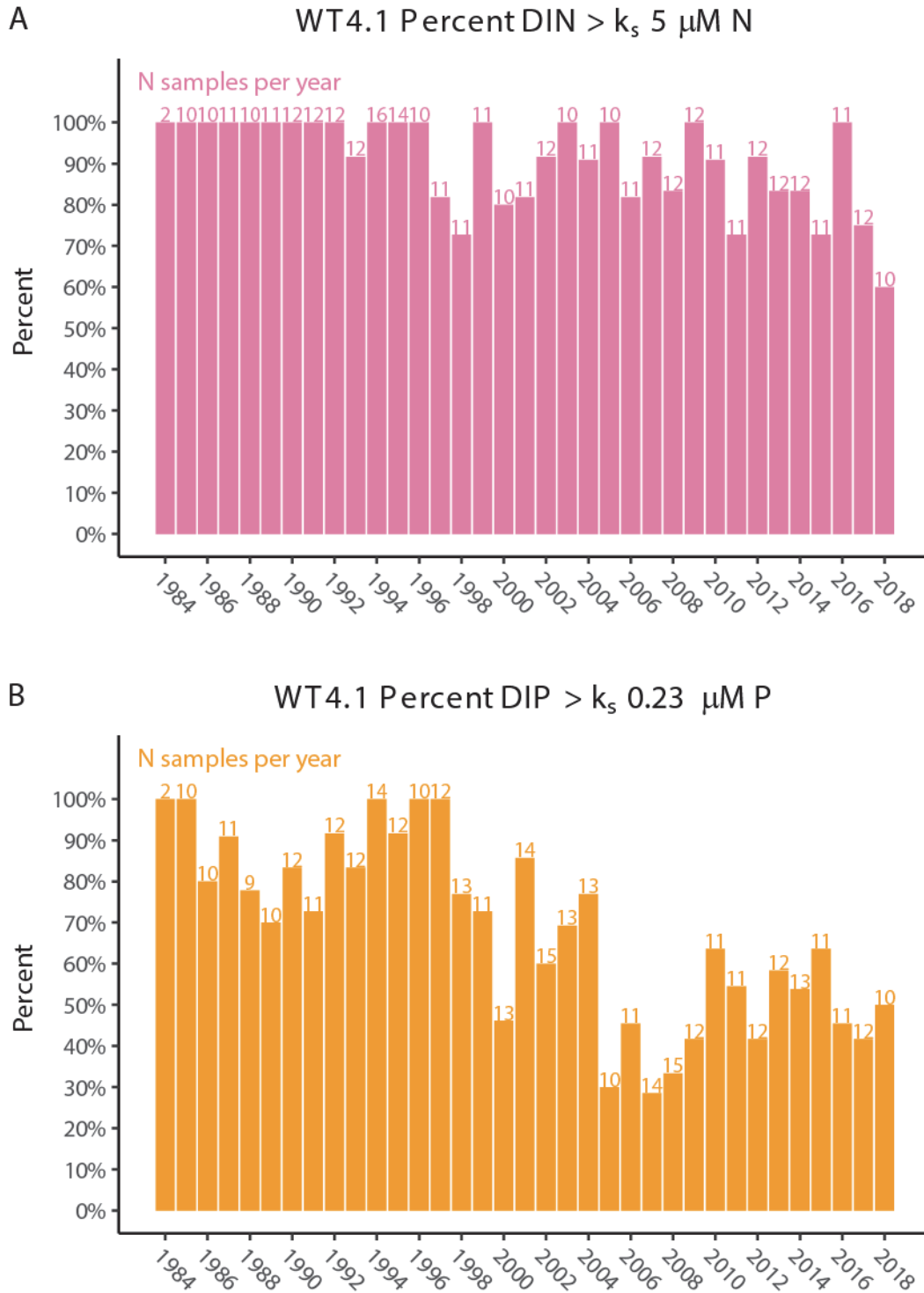
Figure 4. Plots of the percentage of surface water measurements at the Back River long-term monitoring station WT4.1 above k_s values for dissolved inorganic nitrogen (panel A) and phosphorous (panel B). Numbers above bars indicate total sample number that frequency computed from.
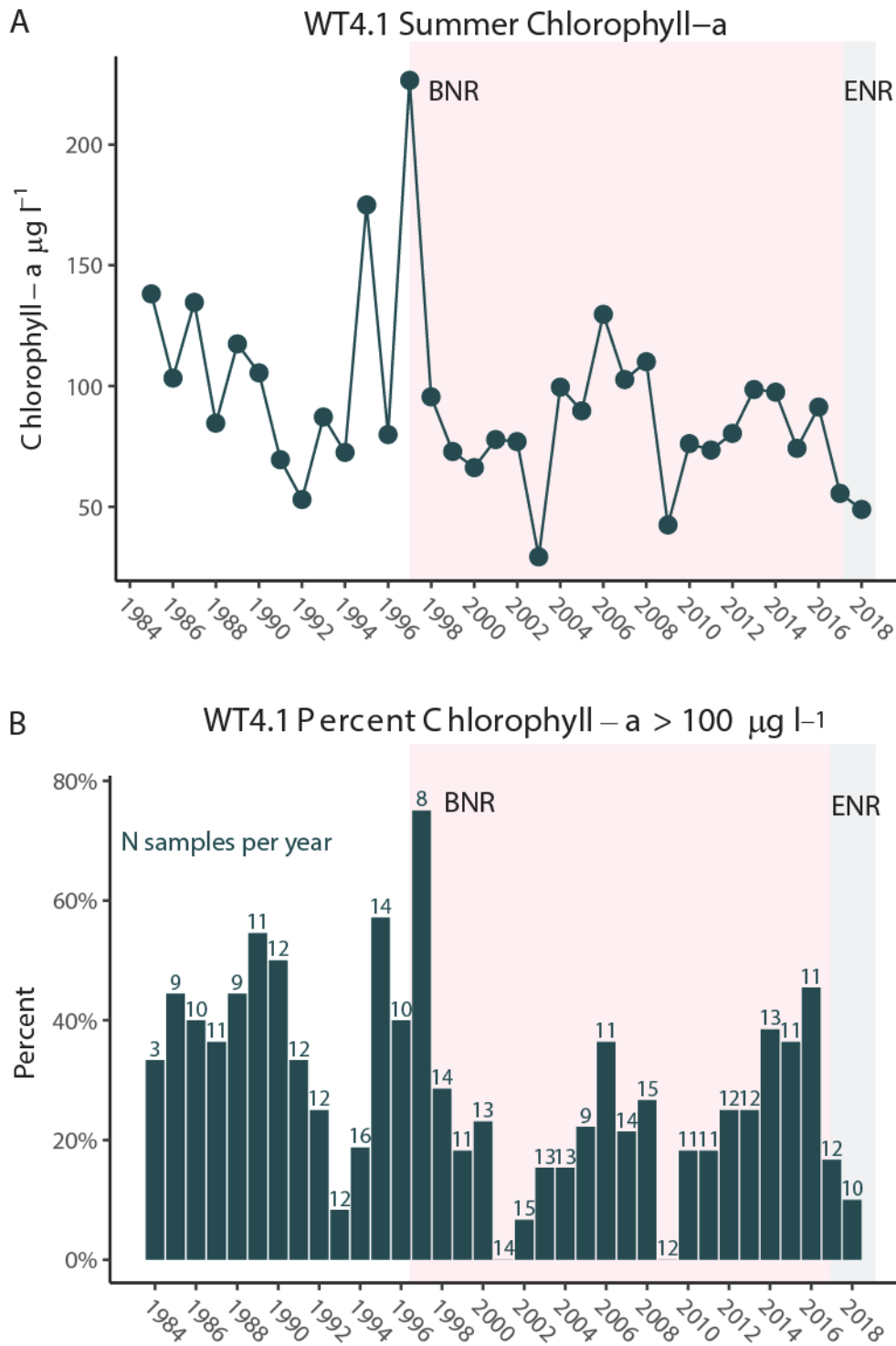
Figure 5. Time-series of average June-August Chlorophyll-*a* (panel A) and the percentage of chlorophyll-*a* measurements greater than 100 μg l^{-1} (panel B). Time periods for biological nutrient removal (BNR) and enhanced nutrient removal (ENR) are shaded on each plot in pink and grey, respectively.
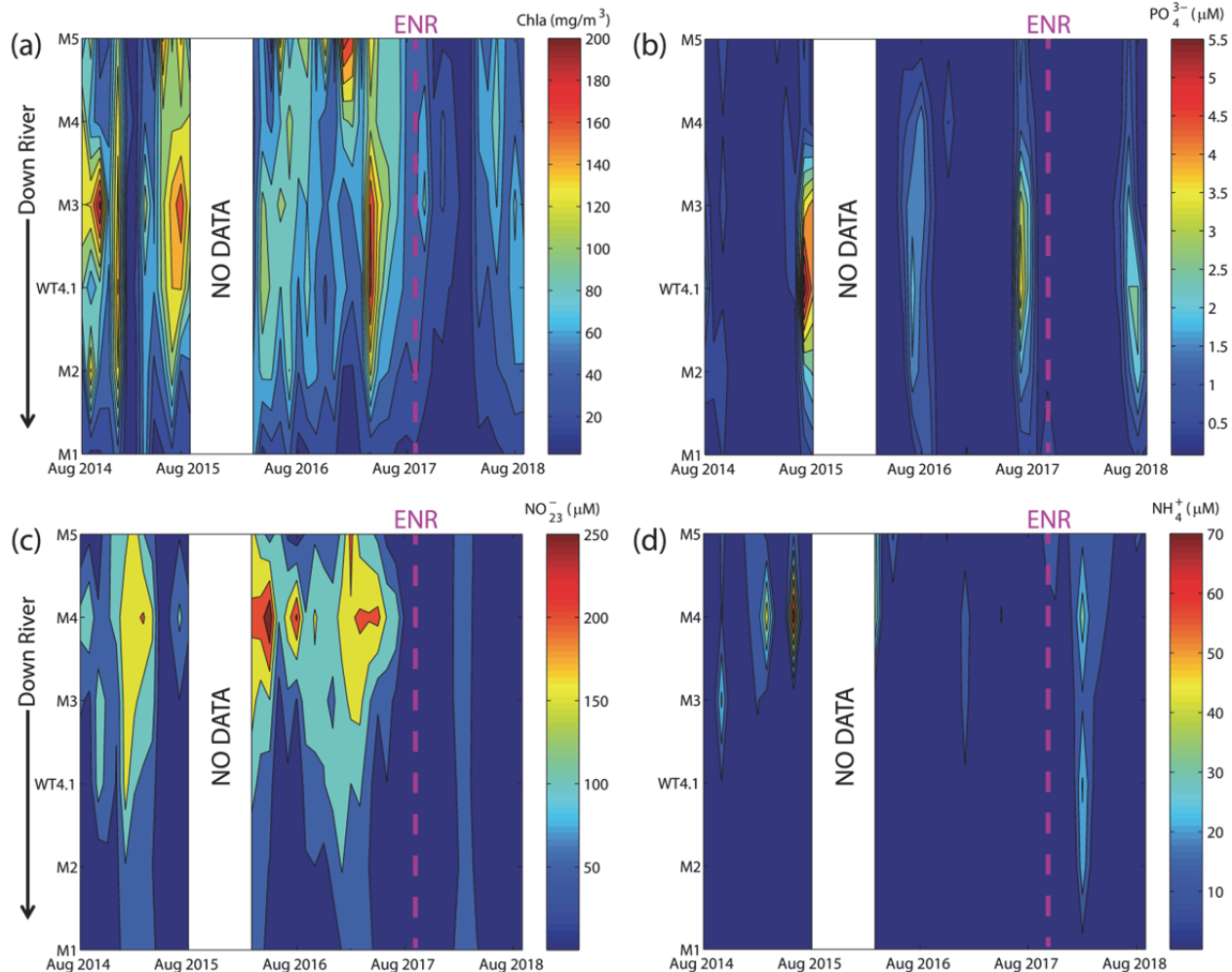
Figure 6. Time-space contour plots of water column parameters at the Maryland Department of the Environment Back River monitoring stations during the period 2014-2018. Data were not collected during a portion of 2015. Clockwise from top left: (a) chlorophyll-a, (b) phosphate (PO$_4$), (c) nitrate+nitrite (NO$_{23}$), and (d) ammonium (NH$_4$).
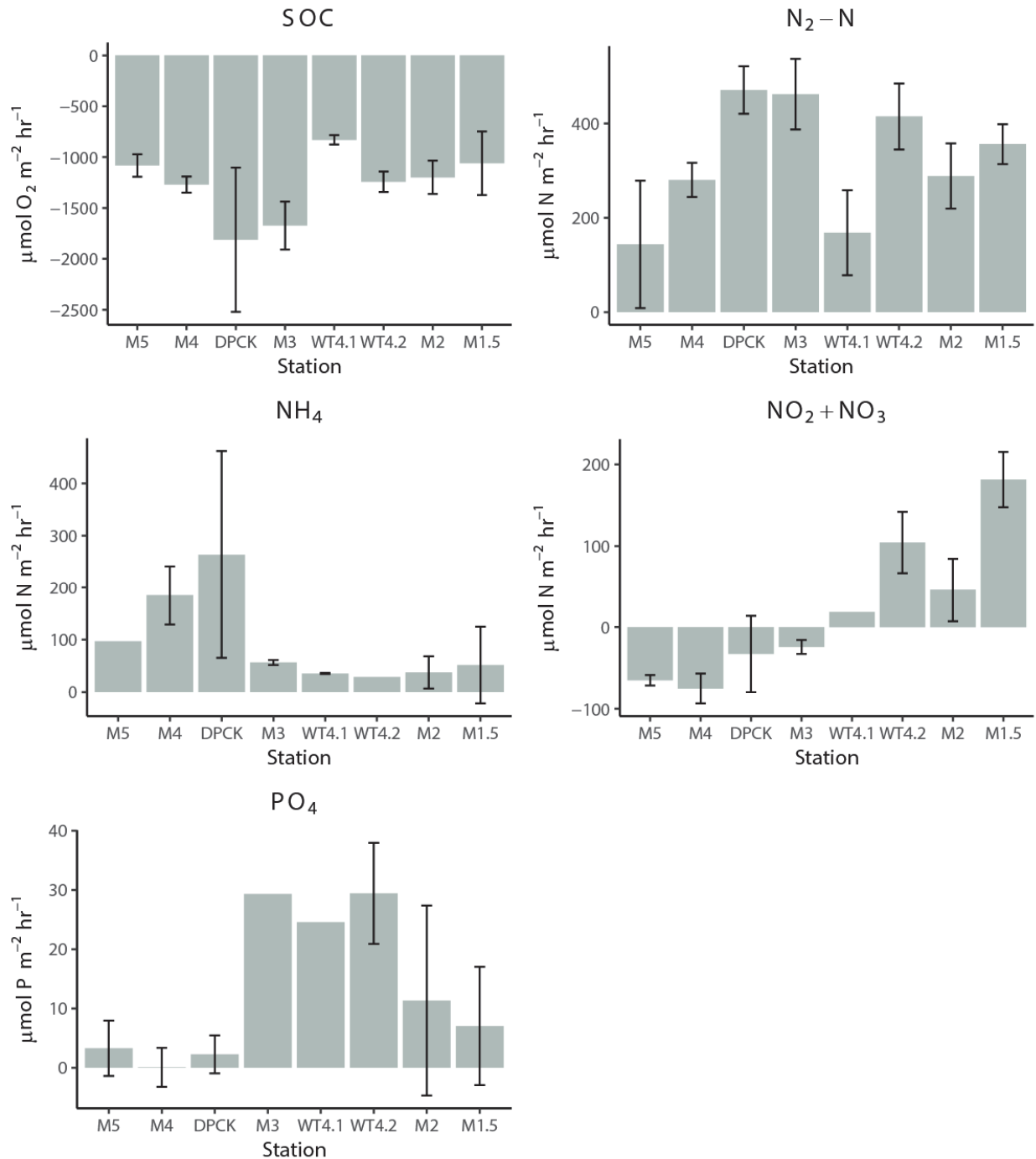
Figure 7. Sediment nutrient and oxygen fluxes measured in the Back River in August, 2018. Bars are oriented from upriver (left) to downriver (right). Error bars represent the standard deviation between replicate cores.
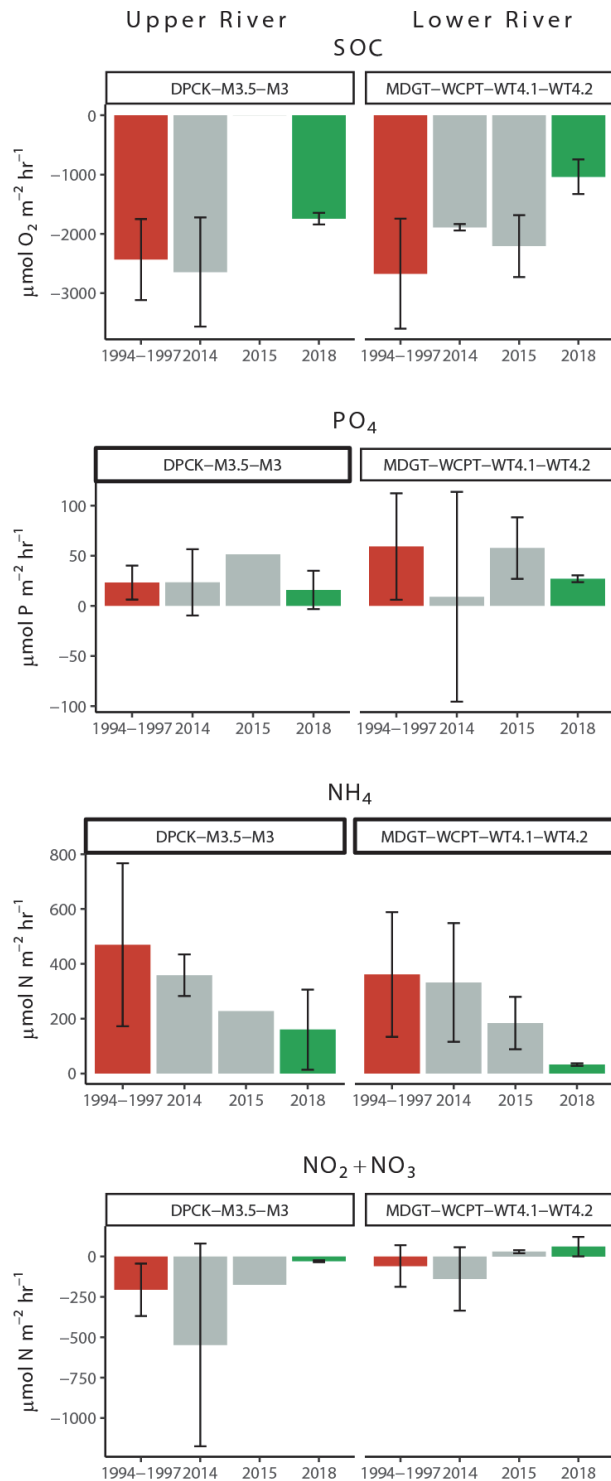
Figure 8. A summary of sediment nutrient and oxygen fluxes measured in the Back River estuary. Left panels are upper river stations and right panels are down river stations (see text for exact stations). Fluxes measured in the 1990s are indicated in red, 2014-2015 fluxes in grey, and 2018 fluxes in green. Error bars represent the standard deviation of the station averages. For ammonium, SOC, and phosphate, sediment-water fluxes in 2018 were significantly lower than the mid-1990s values ($p < 0.05$; Dunn Kruskal-Wallis Multiple Comparison Test).
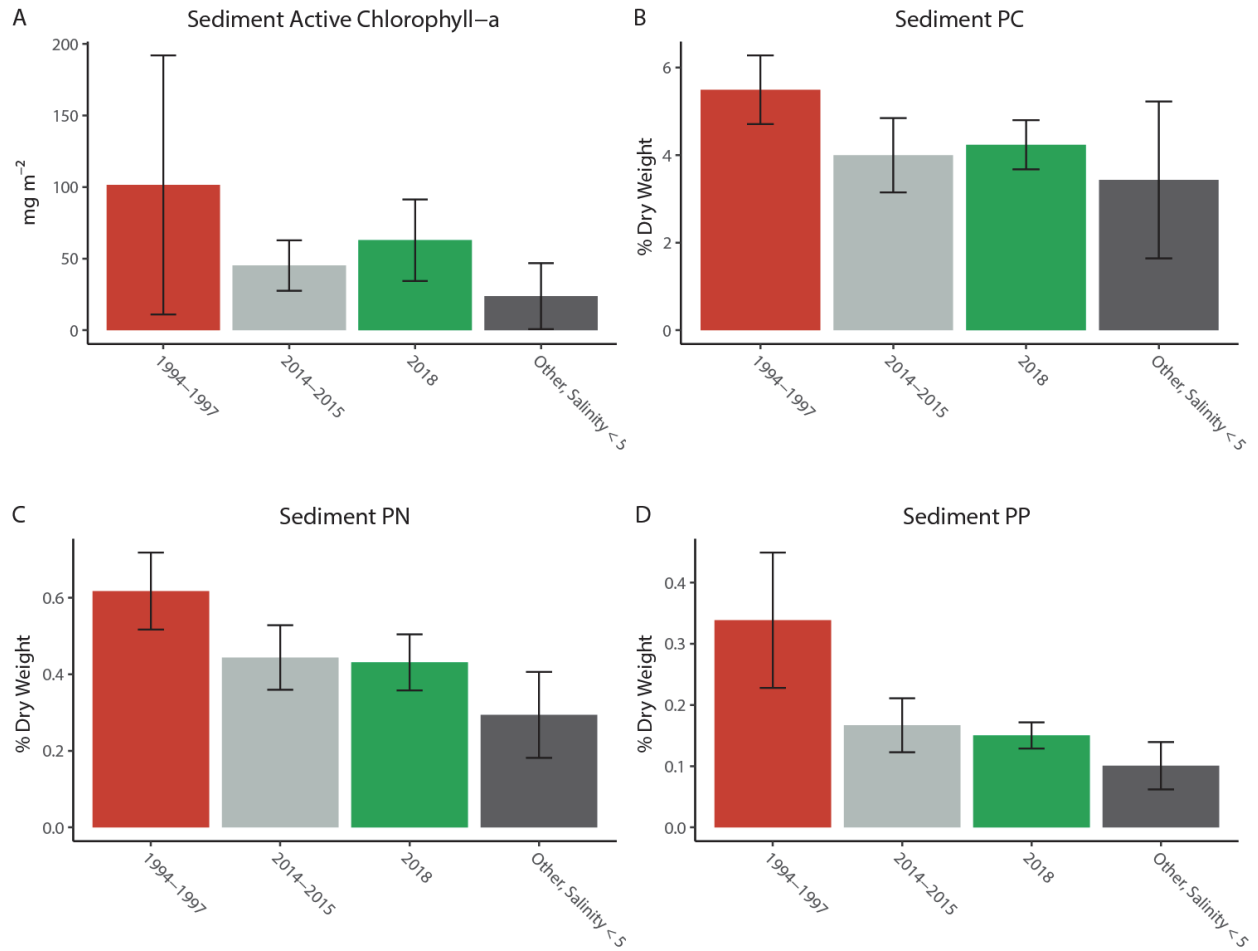
Figure 9. A summary of sediment properties measured in the top 1 cm of sediment cores extracted at the time cores were incubated. Samples measured in the 1990s are indicated in red, 2014-2015 in light grey, 2018 in green. The mean of low-salinity (< 5) Chesapeake Bay stations outside of the Back River are in dark grey. Error bars represent the standard deviation of core averages. For PC, PN, and PP, concentrations in 2014-2015 and 2018 were significantly lower than the mid-1990s values ($p < 0.01$; Dunn Kruskal-Wallis Multiple Comparison Test)
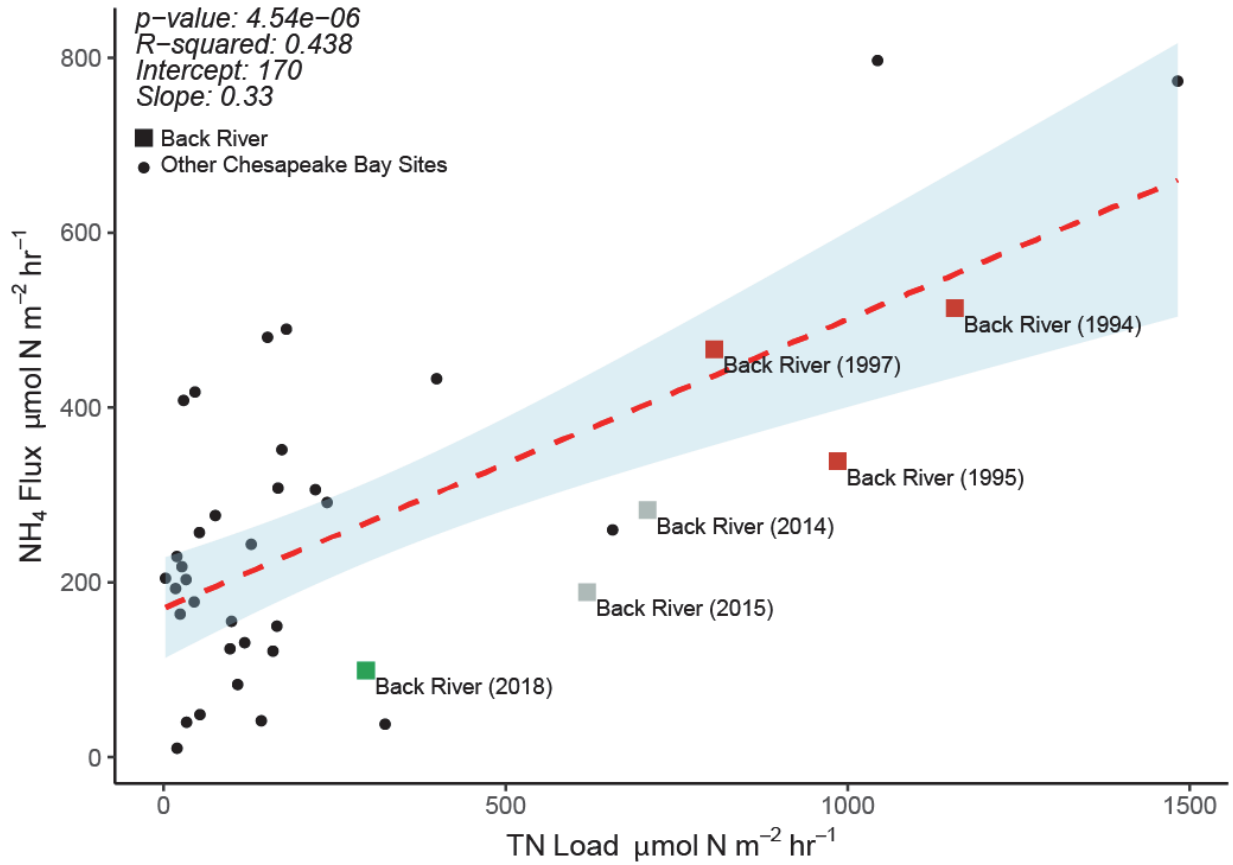
Figure 10. Linear regression of total nitrogen loading rates versus summer (January-August) sediment-water ammonium fluxes (NH_4) in tributaries and the mainstem of Chesapeake Bay (filled circles). Back River data are indicated with squares; 1990s are indicated in red, 2014-2015 in grey, and 2018 fluxes in green, as in Figure 8 & 9.
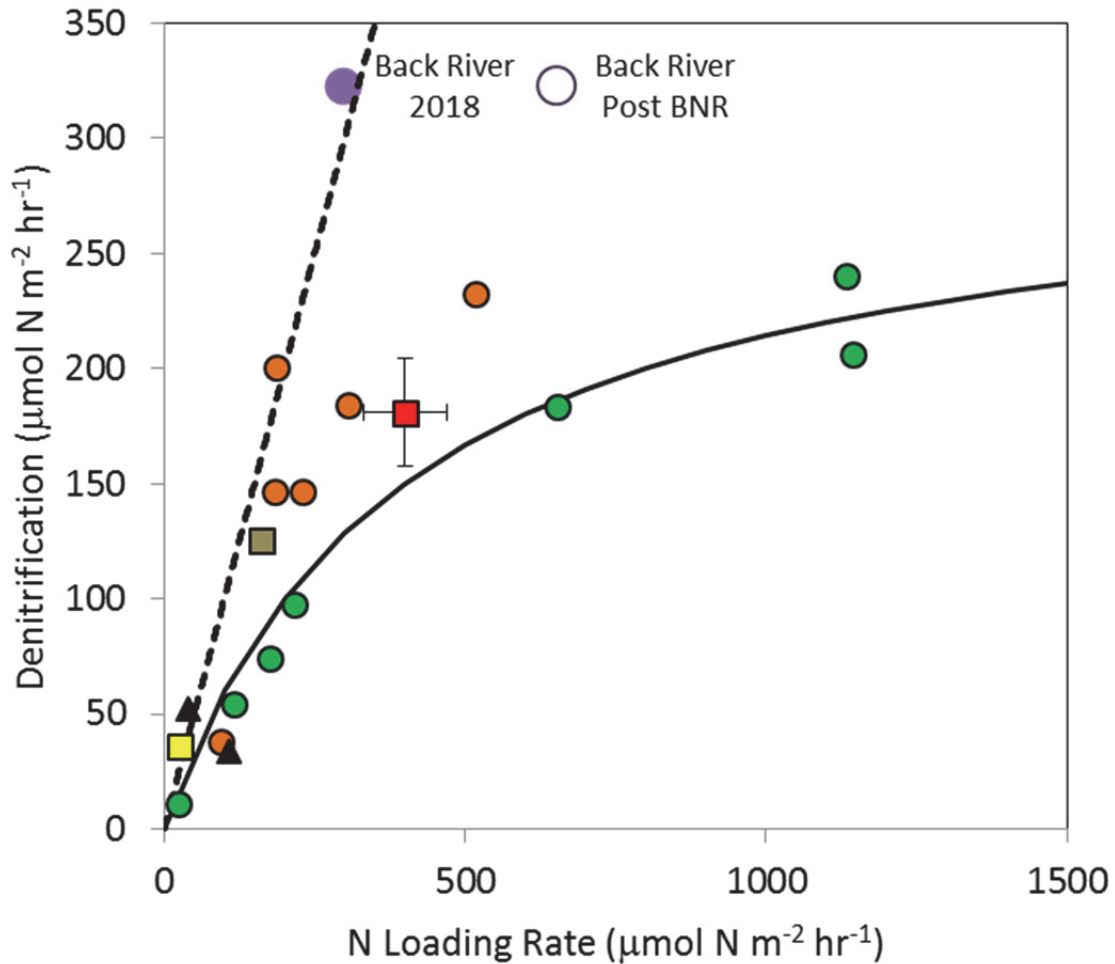
Figure 11. Scatter plot of total nitrogen loading rates versus denitrification rates across a global synthesis of estuarine systems. Green circles from the MERL mesocosms (Oviatt et al. 1996), organge circles from Roskilde Fjord (Kamp-Nelsen 1992), yellow square is Moreton Bay (Eyre and Ferguseon 2009), black triangles are Texas lagoons (Yoon and Benner 1992), and brown square is upper Chesapeake Bay (Testa et al. 2013). Back River denitrification rates from 2018 are related to estimated 2018 loads (filled purple circle) and for loads post-BNR (open purple circle) are included in purple. The solid black line is a best fit function (Denitrification = DN_{mx} * (TN load/(TN Load+ k_{sDN})), where DN_{mx} = 300 and k_{sDN} = 400 µmol m^{-2} h^{-1}, and the dashed line is a 1:1 line, indicating equivalence between denitrification and total nitrogen load.