



# Beneath the surface: spatial and temporal trends in water quality and its impacts on algal community composition in the Albemarle Sound, North Carolina

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**Abstract** Urban and agricultural expansion and intensification pose a critical threat to water quality and aquatic ecosystems. Increased nutrient loading into waterways combined with warming temperatures due to climate change have increased eutrophication and algal blooms. The relationship between land use, nutrient availability, and algal growth can vary dramatically across space and time, but few studies have captured this variation. The goal of this research is to assess water quality across time and disparate land uses, and its influence on algal community composition in the Albemarle Sound, a brackish water estuary in North Carolina. We collected water quality data from 21 sites across the sound, visiting six sites in Chowan County biweekly and 15 other sites twice between June and August 2020. Water samples from

each site were tested for nitrate, phosphate, ammonia, bicarbonate, and total phosphorus (TP). Preserved algal samples from the six Chowan County sites were enumerated under a microscope to estimate genus richness and biomass. In the Chowan County sites, phosphorus increased and nitrate decreased over the course of the summer. Across all sites, TP increased with development and agricultural land use. These results suggest that sources of nitrogen and phosphorus in the sound differ. Algal richness increased with nitrate concentration and decreased with precipitation while biomass increased with water temperature. Our results indicate that climate change impacts, particularly increasing temperatures and extreme precipitation, influence how land use, water quality, and algal community composition interact. These data demonstrate the co-benefits of mitigating climate change in developing management strategies to reduce algal blooms.

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## Introduction

Urban and agricultural expansion pose a critical threat to water quality and aquatic ecosystems. Due to increased nutrient loading from these land uses, harmful algal blooms (HABs) have become increasingly prevalent (Carpenter et al. 1998; Glibert et al.

2014; Paerl and Otten 2013). Patterns associated with climate change, namely warming temperatures and more intense and frequent precipitation, have also exacerbated harmful algal blooms by fostering ideal conditions for algal growth and increasing nutrient pollution into waterways during heavy precipitation (Paerl and Otten 2013). However, it is often unclear which nutrients and other water quality variables drive algal growth. Elucidating the underlying causes of these blooms is crucial to mitigating their impact on humans and natural ecosystems.

Land use types within a watershed can impact the quality of the water it drains. In the USA, most nitrogen (*N*) and phosphorus (*P*) runoff into surface water comes from nonpoint sources of pollution (Carpenter et al. 1998). Agriculture is a major nonpoint pollution source as fertilizers contain *N* and *P* compounds (Carpenter et al. 1998; Glibert et al. 2014; Huang et al. 2017; Le et al. 2010). Nutrients like inorganic *N* are often lost from agricultural fields through leaching or runoff into surface water, increasing nutrient levels downstream (Galloway et al. 2003, Glibert et al. 2014). Urban and suburban land can also influence nutrient runoff (Carpenter et al. 1998; Hobbie et al. 2017; Nielson and Smith 2005). For example, lawn fertilizers comprise a large portion of *N* loading in urban watersheds while pet waste is a major source of *P* (Hobbie et al. 2017). Impervious land area impacts nutrient loading by preventing stormwater from permeating into soil, therefore increasing runoff and nutrient leaching into waterways (Brush 2009; Liu et al. 2013; Xian et al. 2007).

Excess nutrient loading into water bodies can cause algal blooms (Fig. S1, Conley et al. 2009; Galloway et al. 2003; Glibert et al. 2005; Paerl and Otten 2013). Algal blooms pose a severe threat to aquatic ecosystems, with potential consequences for human health. Blooms can result in dead zones and fish kills (Moorman et al. 2014; Paerl and Otten 2013; Watson et al. 2016). Eutrophication and algal blooms may also increase water turbidity, which can negatively impact photosynthetic aquatic vegetation and estuarine predators, further limiting biodiversity (Lunt and Smee 2020; Paerl and Otten 2013, Sukenik et al. 2015). Additionally, some species of blue-green algae (cyanobacteria) secrete toxins like microcystins, a type of cyanotoxin which is connected to a range of health issues in humans, including rashes, liver failure, and cancer (Cheung et al.

2013; Lopez et al. 2008; Paerl 2008; Vranješ and Jovanović, 2011). Dinoflagellates, another type of plankton, also commonly produce toxins and form HABs (e.g., red tide; Fleming et al. 2011). To better protect aquatic ecosystems and human health from this threat, the causes and effects of algal blooms must be explored further.

It was historically assumed that algae in freshwater environments were limited by the availability of *P* (Schindler 1977) and brackish and coastal waters were predominantly *N*-limited. However, mounting evidence suggests that algae are more often co-limited by the availability of both *N* and *P* (Bratt et al. 2019; Elser et al. 2007; Harpole et al. 2011). Beyond evidence of co-limitation, within communities the availability of *N* and *P* can have varying effects on primary producer growth. Non-*N*-fixing genera thrive in water with higher *N*:*P* ratios compared to *N*-fixers (Berry et al. 2017; Paerl and Otten 2013). Furthermore, some species of algae can respond to changing nutrient availability in their environments. For example, Harke et al. (2016) found that *P*-scavenging gene expression in *Microcystis*, a non-*N*-fixer, increased when *P* concentration was low in Lake Erie, giving this genus an advantage over *N*-fixing algae when phosphate concentrations are low. Other species of microalgae in addition to *Microcystis* can uptake excess phosphate and store it in specialized cells (Powell et al. 2011). Thus, while nutrient availability influences algal growth, algae can impact nutrient concentrations through uptake and *N*-fixation. However, nutrient availability itself can vary dramatically across space and time due to factors including human activity, yet few studies have captured this spatiotemporal variation.

The goal of this research is to explore the relationship between space, time, land use, water quality, and algal community composition in the Albemarle Sound region of North Carolina. While much research on water quality and algal blooms has been conducted in the Neuse River Estuary (e.g., Cira et al. 2016; Paerl et al. 1998, 2004, 2010, Pamlico Sound (e.g., Bales 2003; Christian et al. 2004; Paerl et al. 2001, 2007; Piehler et al. 2004), and entire Albemarle-Pamlico Estuary (e.g., Lin et al. 2007; Tester et al. 2003), little research has focused specifically on the Albemarle Sound (e.g., Hazen and Esch 1983; Lombardo 2018; Moorman et al. 2014; Zhang et al. 2014). Thus, the Albemarle Sound is an understudied region despite its

ecological importance as a habitat for many aquatic species (APNEP, n.d.-a, APNEP, n.d.-b).

This research seeks to answer the following questions: (1) How does land use influence water quality in the Albemarle Sound? (2) How does water quality vary across space and time? (3) How does water quality relate to algal taxa richness and biomass? Understanding the interaction between land use and water and how algal communities respond to these factors is crucial to determining potential management strategies and protecting an important estuary and the people who live near it.

## Methods

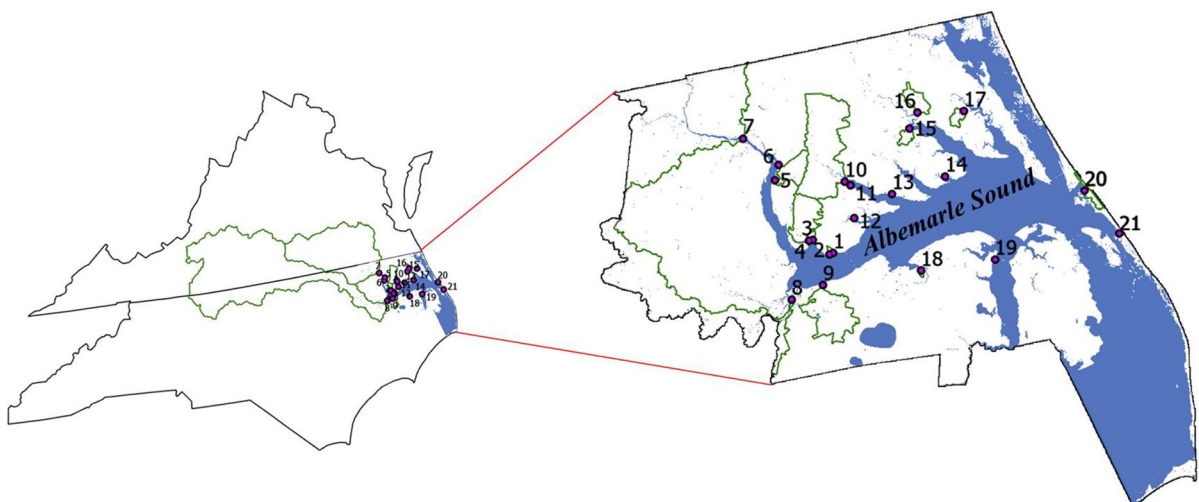
### Study area

The Albemarle Sound (Fig. 1) is part of the Albemarle-Pamlico estuarine system. Contained by barrier islands, it is the second largest estuary in the USA. (APNEP, n.d.-c). The estuary is composed of a diverse assemblage of aquatic biota, such as blue crabs, shrimp, oysters, and several fish species, including Atlantic Sturgeon which rely on the estuary to breed (APNEP, n.d.-a; APNEP, n.d.-b). The sound also provides economic services to northeastern North Carolina including commercial fishing

and aquaculture value as well as recreational value (Van Houtven et al. 2016). The Chowan, Pasquotank, and Roanoke river basins, which extend into Virginia, feed into the sound (APNEP, n.d.-c). The region is threatened by climate change and anthropogenic activity. The rate of relative sea level rise has increased, with potential negative impacts on the estuary ecosystem (Bhattachan et al. 2018; Kemp et al. 2009). Furthermore, agriculture and development in the region have contributed to eutrophication, erosion, and loss of wetlands (APNEP, n.d.-c). The region regularly experiences algal blooms in the summer months (Plaas et al. 2022).

### Field sampling

Field sampling took place over 10 weeks in Chowan County, NC, and 7 other nearby counties (Fig. 1) during summer 2020. Sampling locations were based on public access points and selected to include multiple land use types (e.g., agricultural, developed, wetland) and different locations along an upstream–downstream gradient. Each site in Chowan County ( $n=6$ ) was monitored biweekly (every two weeks), while all other sites ( $n=15$ ) were monitored once a month (a total of 2 times) to capture both spatial and temporal variation. Secchi depth was measured as a proxy for turbidity and a probe was used to determine dissolved



**Fig. 1** Map of study region. Sampling was conducted at 21 sites across 8 counties in Northeastern North Carolina. Sites in Chowan County (1–6) were sampled every two weeks from June 8th to August 10th for a total of five times while the

remaining sites (7–21) were each sampled twice across June and July. The watersheds for each site are outlined in green. We obtained state outlines from the USA Census Bureau (2018) and county outlines from the NCDEQ (2019)

oxygen concentration (Hanna Instruments, HI9142). pH, total dissolved solids (TDS), conductivity (as a proxy for salinity), and temperature were measured on site using a Hanna Instruments Combo (HI98129). Water samples were collected at 0.25 m depth. Weather data were collected from the North Carolina Climate Retrieval and Observations Network of the Southeast Database (North Carolina Climate Office 2020).

### Laboratory analysis

Water samples were filtered using 0.45- $\mu\text{m}$  nylon syringe filters and colorimetrically analyzed for nitrate ( $\text{NO}_3^-$ , cadmium reduction method), phosphate ( $\text{PO}_4$ , ascorbic acid reduction method), and ammonia ( $\text{NH}_3$ , nesslerization method) using a LaMotte Smart 3 colorimeter. Bicarbonate ( $\text{HCO}_3^-$ ) concentration was measured using a LaMotte titration kit (Hardness Titration Reagent). Phosphate was analyzed using a vanadomolybdophosphoric acid method initially for 10 of the study sites. This method was unable to detect phosphate levels, so those measurements were not included in the analysis and an analysis suitable to lower concentrations (ascorbic acid reduction) was used for the remaining samples. Unfiltered water samples from all sites (excluding the first samples from 10 study sites) were frozen and later tested for total phosphorus (TP, ascorbic acid reduction with persulfate digestion method) using a LaMotte Smart 3 Colorimeter.

### Algal sample enumeration

Algal samples from the 6 Chowan County sites were preserved with Lugol's solution (with the concentration dependent on the abundance of algae in the sample) and frozen for six to seven months. Samples were then enumerated using a microscope (Martin Microscope Co. 24,321). One milliliter was drawn from each sample and placed in a centrifuge for 1 min at 100% speed (15,000 rpm). The supernatant was poured off and the sample was resuspended with 0.5–2 mL of distilled water and 0.1 mL of sample was analyzed on a Palmer cell. Cell counts were conducted by examining 100 fields of view, except in the case of one sample where 50 fields of view were analyzed. A Whipple grid was used to aid in genus identification by determining cell size. There were five unknown

groups categorizing cells whose genera remained unidentified. These were included in genus richness and biovolume. All cell counts were later standardized to 100 fields of view and 1 mL of original sample.

Genus richness was calculated as the total number of genera identified in each sample. The biovolume of each genus was calculated by multiplying the standardized counts by an average genus biovolume. Following Passy and Legendre (2006), biovolume was used as a proxy for algal biomass. Biovolume was determined using data from the Swedish Meteorological and Hydrological Institute's Nordic Microalgae and Aquatic Protozoa database, phycology data from the Academy of Natural Sciences and Patrick Center for Environmental Research at Drexel University (ANSP, 2001), a diatom biovolume dataset compiled by Leblanc et al. (2012), and other resources (García et al. 2007; Harrison et al. 2015; Hawkins et al. 2005; Olenina et al. 2006; SMHI, n.d.; Università del Salento, n.d.). Because datasets included measurements for several species of many genera, only measurements which were plausible for the samples collected in this study were included in the average biovolume calculation. Biovolumes of unknown groups were calculated based on Hillebrand et al. (1999).

### Statistical analysis

Watersheds were delineated using StreamStats (USGS, n.d.). The influence of land use on each sampling point was determined by clipping the National Land Cover Database (NLCD) (2016) land cover layer (USGS 2016) to each watershed in ArcGIS Pro 2.5.0 (ESRI 2020) and calculating the proportion of each land use: open water, developed (open space, low, medium, and high intensity), barren land, forest (deciduous, evergreen, mixed), shrub/scrub, herbaceous, agriculture (pasture and cropland), and wetland (woody and emergent herbaceous). ArcGIS Pro was further used to calculate site distance to river mouth.

Linear modeling was used to assess how nutrient concentrations in the Chowan County sites varied over the course of the summer. The models that best fit the data for each water quality variable,  $\text{NO}_3^-$ ,  $\text{PO}_4$ ,  $\text{NH}_3$ ,  $\text{HCO}_3^-$ , and TP, were determined using stepwise AIC model selection (Table S1). Proportion developed, agricultural, forested, and wetland area in

the watershed, antecedent precipitation, date, distance to river mouth (km), and site were included in the modeling. Antecedent precipitation was determined by averaging the daily precipitation (mm) from the week prior to when the sample was collected.

Linear modeling was further used to assess relationships of land use to nutrient concentration among all sites. Similar to the temporal analysis of nutrient concentrations at the Chowan sites, models were chosen using stepwise AIC model selection, taking into account proportion developed, agricultural, forested, wetland area, and distance to river mouth (km; Table S2). Nutrient concentration data were averaged for each site. Because not all sites were visited consistently across time, date and precipitation were excluded from this analysis. Site was also removed as the 21 separate sites created too many variables for the model to run. All statistical analyses were conducted in RStudio version 3.6.2 (RStudio Team 2019).

Linear modeling was also used to determine the relationship between nutrient concentration and algal genus richness, biomass, and percent total biomass of cyanobacteria, diatoms, and dinoflagellates (Table S3, Table S4, Table S5). For genus richness and biomass, AIC model selection was used to find the best model among the following variables:  $\text{NO}_3^-$ ,  $\text{PO}_4$ ,  $\text{NH}_3$ , and  $\text{HCO}_3^-$  concentration, precipitation, site name, date, proportion forest, agriculture, wetland, and developed land, distance from river mouth, water temperature, Secchi depth, pH, conductivity, and TDS. Total phosphorus could not be included in model selection as TP was not collected for the first two samples (sites 5 and 6; June 13). Percent algal biomass was calculated for each genus in each sample to discern any differences in which algae types were dominant between sites. To assess the influence of nutrient concentrations, and water temperature and salinity on the proportion total biomass of cyanobacteria, diatoms, and dinoflagellates, stepwise AIC model selection for the percent biomass of those algal groups included  $\text{NO}_3^-$ ,  $\text{PO}_4$ ,  $\text{NH}_3$ , and  $\text{HCO}_3^-$  concentrations, water temperature, site, and conductivity.

## Results

Watersheds varied in size and land use, with development, agriculture, forest, and wetland-dominated

watersheds. Agriculture was the most predominant land use in study sites' watersheds, followed by wetland, developed land, and forest (Table 1). Agriculture composed the greatest proportion of land area in 11 sites, while wetland, forest, and developed land made up the greatest proportion of land area in 4, 3, and 3 sites, respectively. Algal blooms were observed at some sites during the study period, two of which had watersheds dominated by agricultural land use while another had a predominantly forested watershed (Fig. S2).

### Temporal variation in water quality

The linear model results for nutrient levels at the six Chowan County sites visited biweekly demonstrate that nitrate and phosphorus levels both changed over time, but in different ways (Table 2).  $\text{NO}_3^-$  decreased significantly over time ( $p < 0.05$ ) while TP ( $p < 0.001$ )

**Table 1** Land use proportions for study site watersheds. Watersheds were delineated using StreamStats (USGS, n.d.) and land use proportions were obtained using the NLCD 2016 land cover layer (USGS, 2016). Multiple development, agriculture, forest, and wetland types were combined to obtain the total proportions of each land use

Site	%Developed	%Agriculture	%Forest	%Wetland
1	61.32	2.32	15.21	6.50
2	61.32	20.28	10.85	1.42
3	7.66	41.84	30.48	16.12
4	4.88	43.32	32.39	15.88
5	4.59	54.15	17.25	21.04
6	11.37	55.61	9.68	23.20
7	4.98	20.50	48.46	14.18
8	7.57	20.39	56.56	4.79
9	4.13	47.85	12.60	31.42
10	3.35	45.34	13.40	35.54
11	4.76	78.91	0.00	6.12
12	17.46	29.37	3.97	48.41
13	1.14	12.50	48.86	36.36
14	1.42	85.79	3.02	9.41
15	36.59	50.56	4.96	7.51
16	5.95	68.10	4.87	20.56
17	3.26	48.93	7.92	38.95
18	11.76	25.34	0.77	61.51
19	0.00	18.39	0.00	81.61
20	72.47	0.24	7.55	5.26
21	18.68	0.36	0.00	67.62

**Table 2** Results of linear models for temporal analysis of Chowan County sites, visited biweekly.

Response variable	AIC	R <sup>2</sup>	Explanatory variables	Coefficient	Pr(> t )	95% CI
Avg. nitrate (NO <sub>3</sub> <sup>-</sup> , ug/L)	283.9	0.48	Intercept	44,202.124	0.014 *	9657.363, 78,746.880
			<b>site 1</b>	<b>-177.686</b>	<b>0.012 *</b>	<b>-312.209, -43.162</b>
			<b>site 2</b>	<b>-209.686</b>	<b>0.004 **</b>	<b>-344.209, -75.162</b>
			<b>site 3</b>	<b>-193.966</b>	<b>0.007 **</b>	<b>-328.290, -59.643</b>
			<b>site 4</b>	<b>-203.243</b>	<b>0.005 **</b>	<b>-337.578, -68.908</b>
			site 6	-105.636	0.117	-239.912, 28.640
			<b>date</b>	<b>-2.383</b>	<b>0.015 *</b>	<b>-4.255 -0.5112</b>
Total phosphorus (TP, ug/L)	249.32	0.81	Intercept	-145,800.000	<0.001 ***	-222,590.700, -69,020.940
			<b>precipitation (mm)</b>	<b>31.200</b>	<b>&lt;0.001 ***</b>	<b>18.213, 44.193</b>
			site 1	-202.000	0.092	-441.018, 37.086
			site 2	-137.000	0.242	-376.018, 102.086
			site 3	-115.300	0.323	-354.725, 124.217
			site 4	78.330	0.494	-159.045, 315.712
			site 6	-225.300	0.064	-464.869, 14.290
<b>date</b>	<b>7.908</b>	<b>&lt;0.001 ***</b>	<b>3.747, 12.069</b>			
Avg. phosphate (PO <sub>4</sub> , ug/L)	286.47	0.68	Intercept	-48,240.928	0.025 *	-89,963.000, -6518.859
			precipitation (mm)	5.690	0.145	-2.126, 13.506
			site 1	-37.360	0.584	-176.857, 102.137
			site 2	-35.360	0.604	-174.857, 104.137
			site 3	-2.018	0.977	-142.165, 138.130
			<b>site 4</b>	<b>248.764</b>	<b>0.001 **</b>	<b>109.579, 387.950</b>
			site 6	-59.877	0.384	-199.783, 80.029
<b>date</b>	<b>2.616</b>	<b>0.025 *</b>	<b>0.354, 4.879</b>			
Avg. ammonia (NH <sub>3</sub> , ug/L)	287.55	0.47	Intercept	402.400	2.26e-08 ***	300.512, 504.288
			<b>site 1</b>	<b>-237.600</b>	<b>0.002 **</b>	<b>-381.692, -93.508</b>
			<b>site 2</b>	<b>-206.800</b>	<b>0.007 **</b>	<b>-350.892, -62.708</b>
			<b>site 3</b>	<b>-184.800</b>	<b>0.014 *</b>	<b>-328.892, -40.708</b>
			site 4	-54.400	0.444	-198.492, 89.692
			site 6	-33.600	0.635	-177.692, 110.492
			<b>date</b>	<b>2.616</b>	<b>0.025 *</b>	<b>0.354, 4.879</b>
Avg. bicarbonate (HCO <sub>3</sub> <sup>-</sup> , ug/L)	568.22	0.42	Intercept	2,510,255.800	0.201	-1,437,931.000, 6,458,442.000
			<b>site 1</b>	<b>16,816.800</b>	<b>0.033 *</b>	<b>1441.844, 32,191.820</b>
			site 2	14,660.800	0.061	-714.156, 30,035.820
			site 3	7540.200	0.320	-7811.909, 22,892.300
			<b>site 4</b>	<b>15,687.400</b>	<b>0.046 *</b>	<b>334.059, 31,040.770</b>
			site 6	-3701.700	0.623	-19,048.340, 11,645.020
			date	-133.900	0.208	-347.853, 80.049

Table includes the explanatory variables, AIC and R<sup>2</sup> values for each model, coefficients, and 95% confidence intervals of each best fit model. Models were chosen using stepwise AIC model selection. Significant variables (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ) are in bold

and PO<sub>4</sub> ( $p < 0.05$ ) increased over the course of the summer. There was also a significant, positive association between TP and precipitation ( $p < 0.001$ ). While PO<sub>4</sub> concentrations also increased with precipitation, this relationship was not significant ( $p > 0.05$ ). Time did not influence NH<sub>3</sub> concentration. HCO<sub>3</sub><sup>-</sup> similarly

did not vary significantly over time. Both NH<sub>3</sub> and HCO<sub>3</sub><sup>-</sup> concentrations were predominantly influenced by differences between sites (Fig. S3).

While the model chosen through stepwise AIC model selection did not include precipitation as a variable influencing NO<sub>3</sub><sup>-</sup>, the general negative trend

in nitrate concentration corresponded with a spike in rainfall at the end of the summer during Hurricane Isaias (Fig. 2). While TP concentrations were highest at most sites on the last day of sample collection, the  $\text{NO}_3^-$  levels at most sites were lower than previously observed (Fig. 2).

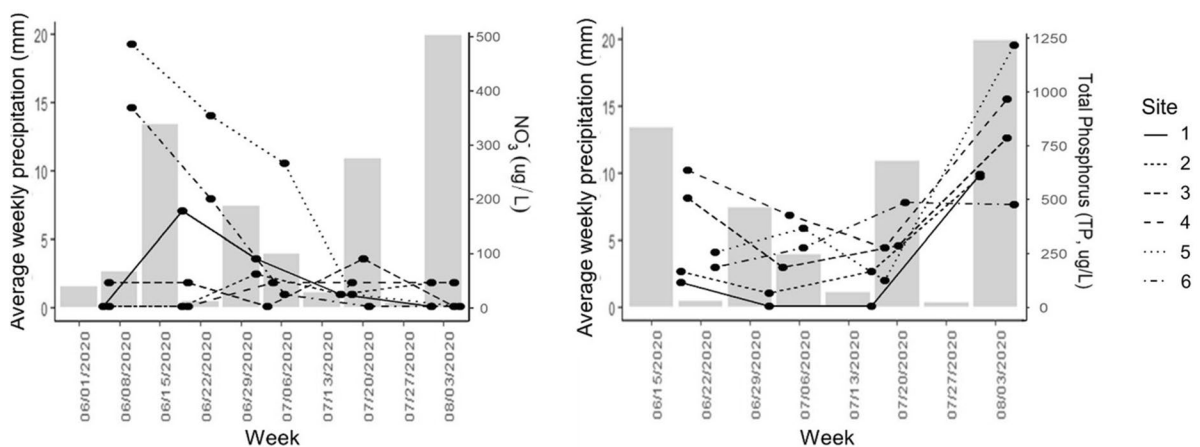
### Spatial variation in water quality

The linear modeling results for average nutrient concentrations across all study sites revealed that TP was the only nutrient measured that was significantly impacted by land use (Table 3). TP increased with increasing proportions of developed ( $p < 0.05$ ) and agricultural ( $p < 0.05$ ) land use in the watershed (Fig. 3). Both  $\text{PO}_4$  ( $p < 0.05$ ) and  $\text{NH}_3$  ( $p < 0.05$ ) exhibited significant, positive relationships with distance to river mouth, meaning sites farther downstream tended to have lower  $\text{PO}_4$  and  $\text{NH}_3$  levels (Fig. S4).  $\text{HCO}_3^-$  was negatively associated with proportion of developed land, farmland, forest, and wetland, as well as distance to river mouth, but these variables were not statistically significant (Table 3). The best fit model for  $\text{NO}_3^-$  did not include any explanatory variables (Table 3).

### Variation in algal community composition

A total of 65 algal genera (12 Cyanobacteria, 16 Chlorophyta, 1 Charophyta, 3 Euglenozoa, 23 Bacillariophyceae (diatoms), and 10 Dinoflagellata) plus 5 unknown groups were identified in 26 samples from the six Chowan County sites (Table S6). Overall, the most abundant genera based on standardized cell counts (rounded to nearest whole number) were *Microcystis* ( $n = 9250$ ), *Nodularia* ( $n = 2077$ ), and *Anabaena* ( $n = 1402$ ), while the genera with the greatest total biomass were *Microcystis* ( $3.2 \times 10^5 \mu\text{m}^3$ ), *Gymnodinium* ( $2.4 \times 10^5 \mu\text{m}^3$ ), and *Nodularia* ( $2.6 \times 10^5 \mu\text{m}^3$ ; Table S6). Twenty-three (35.4%) of the identified genera have confirmed or potential toxic strains, of which 11 (47.8%) were Cyanobacteria and nine (39.1%) were dinoflagellates (Table S6). Six (9.2%) genera can fix atmospheric nitrogen, all Cyanobacteria (Table S6).

Algal genus richness ranged from seven to 38 genera. The sites with the greatest mean richness were sites 3 and 4, while sites 5 and 6 had the lowest average genus richness, but the greatest average biomass (Table S7). Sites 1 and 2 had the lowest average biomass (Table S7). Total standardized biomass ranged from a minimum of  $1,015.05 \mu\text{m}^3$  to a maximum of  $420,529.58 \mu\text{m}^3$  in one sample. However, differences in genus richness and total biomass between sites were not statistically significant (Fig. S5).



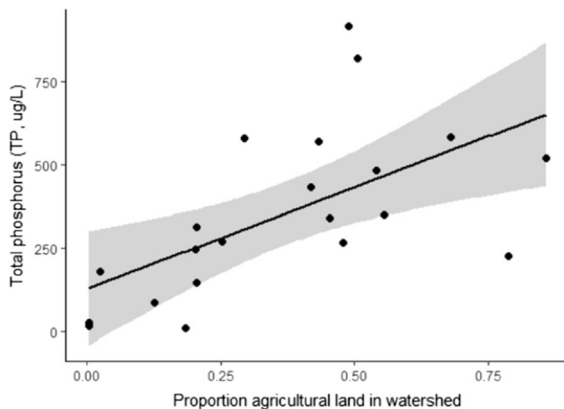
**Fig. 2**  $\text{NO}_3^-$  (left) and TP (right) concentrations and average weekly precipitation. The figures illustrate the  $\text{NO}_3^-$  and TP concentrations at each site and precipitation over the summer. Bars represent average weekly rainfall, patterned lines indicate

different sites ( $n = 6$ ), and points represent visits to each site ( $n = 30$ ;  $n = 24$ ). During the last week, Hurricane Isaias hit the region, causing a dramatic rise in precipitation from the previous week

**Table 3** Results of nutrient concentration and land use linear models for all study sites.

Response variable	AIC	R <sup>2</sup>	Explanatory variables	Coefficient	Pr(> t )	95% CI
Total phosphorus (TP, ug/L)	226.44	0.55	Intercept	-2140.770	0.060	-4380.288, 98.748
			<b>proportion developed</b>	<b>2700.915</b>	<b>0.044 *</b>	<b>90.461, 5311.370</b>
			<b>proportion agriculture</b>	<b>2750.656</b>	<b>0.019 *</b>	<b>510.114, 4991.198</b>
			proportion forest	2472.515	0.056	-76.400, 5021.430
			proportion wetland	2250.499	0.055	-53.195, 4554.193
			distance to river mouth (km)	4.975	0.192	-2.779, 12.729
Avg. nitrate (NO <sub>3</sub> <sup>-</sup> , ug/L)	283.34	N/A	Intercept	392.300	0.043 *	13.877, 770.669
Avg. phosphate (PO <sub>4</sub> , ug/L)	216.68	0.20	Intercept	87.000	0.096	-16.909, 190.910
			<b>distance to river mouth (km)</b>	<b>6.044</b>	<b>0.044 *</b>	<b>0.193, 11.894</b>
Avg. ammonia (NH <sub>3</sub> , ug/L)	233.44	0.43	Intercept	52.594	0.690	-220.836, 326.024
			proportion agriculture	387.786	0.127	-121.628, 897.200
			proportion wetland	400.870	0.119	-114.651, 916.391
			<b>distance to river mouth (km)</b>	<b>9.751</b>	<b>0.038 *</b>	<b>0.619, 18.884</b>
Avg. bicarbonate (HCO <sub>3</sub> <sup>-</sup> , ug/L)	443.47	0.53	Intercept	406,474.500	0.044 *	13,486.310, 799,462.800
			proportion developed	-332,182.600	0.143	-790,262.400, 125,897.200
			proportion agriculture	-350,282.600	0.077	-743,450.500, 42,885.260
			proportion forest	-415,661.500	0.066	-862,942.400, 31,619.300
			proportion wetland	-272,827.400	0.171	-677,077.200, 131,422.300
			distance to river mouth (km)	-933.500	0.164	-2294.116, 427.108

Table includes the explanatory variables, AIC and R<sup>2</sup> values for each model, coefficients, and 95% confidence intervals of each best fit model. Water quality data from every visit were averaged for each site to determine variation across all study sites. Models were chosen using stepwise AIC model selection. Significant variables (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ) are in bold



**Fig. 3** Relationship between proportion of agricultural land and TP. Points represent study sites ( $n=21$ ), best fit line shows the relationship between TP and proportion farmland, and shading indicates a 95% confidence interval. There was a significant, positive association between TP and proportion agricultural land cover in site watersheds among all study sites ( $p < 0.05$ ). See Table 3 for slope based on full model

According to linear modeling results, date appeared to have the strongest influence on genus richness, with richness increasing over time ( $p < 0.001$ ; Table 4). This relationship between algal richness and time held true for each site (Fig. 4). Genus richness was positively related to NO<sub>3</sub><sup>-</sup> ( $p < 0.05$ ) and HCO<sub>3</sub><sup>-</sup> concentration ( $p < 0.01$ ; Table 4). Richness was negatively associated with precipitation ( $p < 0.05$ ) but increased with pH ( $p < 0.01$ ; Table 4).

The best linear model for algal biomass included HCO<sub>3</sub><sup>-</sup> concentration, water temperature, conductivity, and TDS (Table 5). Of these variables, biomass was significantly positively associated with water temperature ( $p < 0.001$ ; Table 5; Fig. 5).

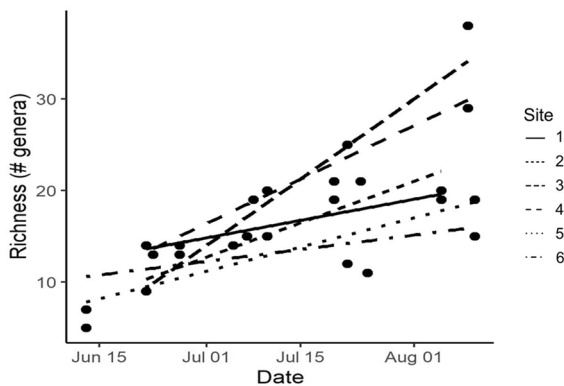
Algal biomass composition (based on percent of total biomass) varied significantly across space and time. Mean proportions of total biomass of cyanobacteria, green algae, diatoms, dinoflagellates, euglenozoan, and unknowns for each site showed that biomass at sites 1 and 2 was dominated by diatoms, mean total biomass at sites 3 and 4 was mainly composed of dinoflagellates, while cyanobacteria largely comprised the mean biomass of sites 5 and



**Table 4** Algal genus richness linear model results.

AIC	$R^2$	Explanatory variables	Coefficient	Pr(> t )	95% CI
58.03	0.94	Intercept	-7885.000	<0.001 ***	-11,113.610, -4656.938
		<b>avg. nitrate (NO<sub>3</sub><sup>-</sup>, ug/L)</b>	<b>0.037</b>	<b>0.024 *</b>	<b>0.006, 0.068</b>
		avg. phosphate (PO <sub>4</sub> , ug/L)	0.013	0.206	-0.009, 0.035
		<b>avg. bicarbonate (HCO<sub>3</sub><sup>-</sup>, ug/L)</b>	<b>0.001</b>	<b>0.004 **</b>	<b>0.0002, 0.001</b>
		<b>precipitation (mm)</b>	<b>-0.482</b>	<b>0.018 *</b>	<b>-0.858, -0.105</b>
		site 1	-9.032	0.119	-20.884, 2.819
		site 2	-9.956	0.687	-22.245, 2.334
		site 3	-2.062	0.697	-13.651, 9.528
		site 4	-2.336	0.186	-15.016, 10.344
		site 6	-4.112	0.100	-10.603, 2.379
		<b>date</b>	<b>0.423</b>	<b>&lt;0.001 ***</b>	<b>0.249, 0.597</b>
		water temperature (°C)	-1.565	0.102	-3.507, 0.378
		Secchi depth (m)	-9.508	0.100	-21.230, 2.215
		<b>pH</b>	<b>1.620</b>	<b>0.005 **</b>	<b>6.241, 26.166</b>
		conductivity (µs)	-0.122	0.302	-0.375, 0.130
		total dissolved solids (TDS)	0.240	0.321	-0.277, 0.758

Table includes the AIC and  $R^2$  values, coefficients, and 95% confidence intervals for the best genus richness model. The model was chosen using stepwise AIC model selection. Significant variables (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ) are in bold



**Fig. 4** Relationship between algal genus richness and time by study site. Points represent visits to each site ( $n=26$ ); color-coded best fit lines represent different sites ( $n=6$ ). The linear modeling results indicate that algal genus richness increased significantly over time ( $p < 0.001$ ), though site was not included in the best model

6 (Fig. S6). While different algal groups appeared to dominate different sites, only the mean percent total biomass of cyanobacteria differed significantly among groups ( $p < 0.01$ , Fig. S7). Algal community composition also shifted over time. The proportion total biomass composed of diatoms at sites 1 and 2 increased over time, peaking in August, while

the proportion of dinoflagellate biomass at sites 3 and 4 was greatest in early July (Fig. 6). The relative biomass of cyanobacteria was overwhelmingly dominant at sites 5 and 6 in late July, with a drastic decline in early August as the relative biomass of dinoflagellates increased (Fig. 6).

Because cyanobacteria, diatoms, and dinoflagellates were the most prevalent algal groups in terms of proportion of total biomass, linear modeling was used to determine the influence of water quality (nutrient concentration, conductivity, and water temperature) on the relative biomass of these three algal groups. The relative biomass of cyanobacteria increased significantly with increasing NO<sub>3</sub><sup>-</sup> concentrations and water temperature while dinoflagellate relative biomass was significantly negatively associated with NO<sub>3</sub><sup>-</sup> (Table 6; Fig. 7). The model selected for diatom relative biomass did not include any of the variables fed into the model.

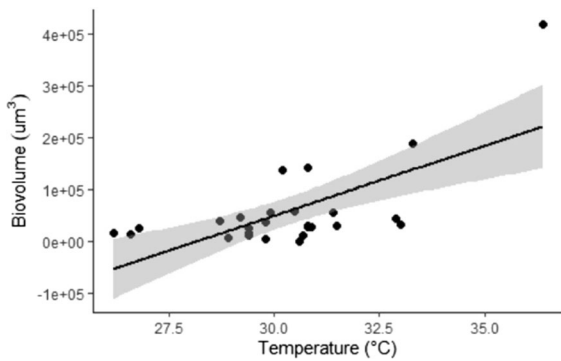
## Discussion

Our results demonstrate that water quality characteristics varied differently across space, time, and land use, with nitrate concentration predominantly driven

**Table 5** Algal biomass (as biovolume) linear model results.

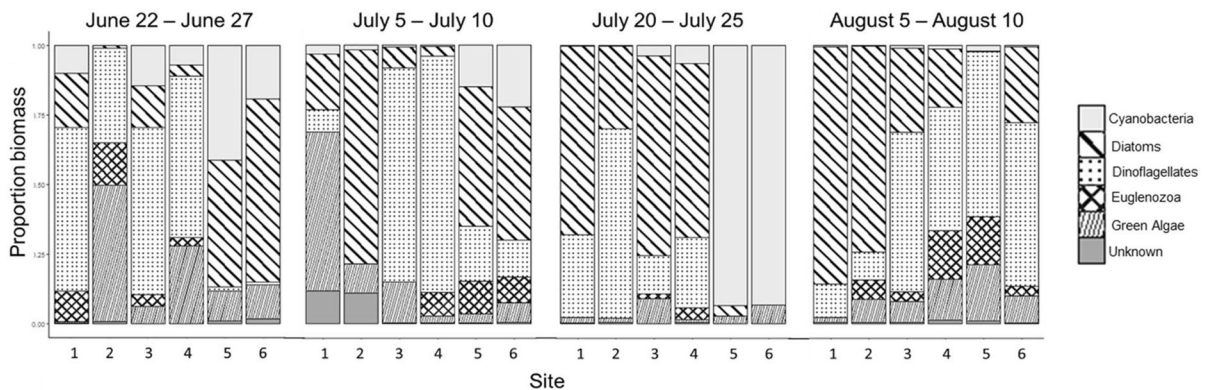
AIC	R <sup>2</sup>	Explanatory variables	Coefficient	Pr(> t )	95% CI
555.65	0.60	Intercept	-740,900.000	<0.001 ***	-1,129,429.000, -352,312.300
		avg. bicarbonate (HCO <sub>3</sub> <sup>-</sup> , ug/L)	-3.4110	0.070	-7.121, 0.300
		<b>water temperature (°C)</b>	<b>30,960.000</b>	<b>&lt;0.001 ***</b>	<b>18,296.060, 43,631.370</b>
		conductivity (µs)	2453.000	0.165	-1100.707, 6006.915
		total dissolved solids (TDS)	-4810.000	0.173	-11,904.570, 2283.638

Table includes the AIC and R<sup>2</sup> values, coefficients, and 95% confidence intervals for the best algal biomass model. The model was chosen using stepwise AIC model selection. Significant variables (\*: p < 0.05, \*\*: p < 0.01, \*\*\*: p < 0.001) are in bold



**Fig. 5** Relationship between total biomass (as biovolume) and water temperature. Points represent visits to each site (n = 26); best fit line represents the relationship between algal biomass and water temperature, and gray shading represents a 95% confidence interval. Algal biomass (measured as biovolume) increased significantly with water temperature (p < 0.001). See Table 5 for slope based on full model

by time and phosphorus concentration driven by time, precipitation, and land use. Analysis of the Chohan County sites (those visited biweekly) revealed that TP increased significantly over time and with precipitation while NO<sub>3</sub><sup>-</sup> decreased over time. Across all 21 sites, TP significantly increased with proportion of agricultural and developed land use. Algal genus richness increased over time and with NO<sub>3</sub><sup>-</sup> concentration but decreased with precipitation. However, algal biomass was significantly positively related to water temperature. Similarly, percent total biomass of specific algal groups varied with NO<sub>3</sub><sup>-</sup> concentration and temperature. We explore these results below.



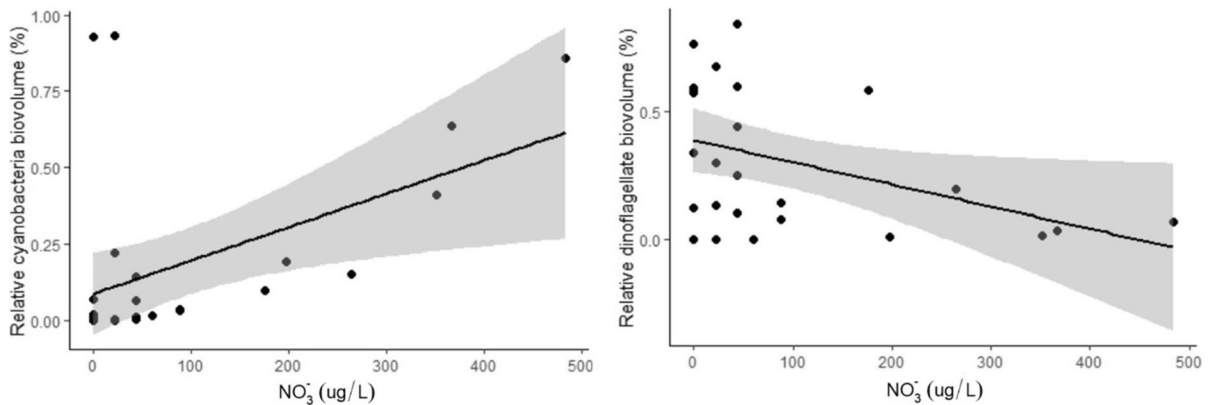
**Fig. 6** Proportion biomass of algal groups including unknowns over time at each site. Stacked bar plots show the proportion biomass (measured as biovolume) of the 5 algal groups identified in this study and unknowns. The first two

algal samples at sites 5 and 6 (collected on June 13th) were omitted from this chart as algal taxa data was not collected for the remaining four sites until the following week

**Table 6** Cyanobacteria, diatoms, and dinoflagellates percent total biomass (as biovolume) linear modeling results.

Response variable	AIC	$R^2$	Explanatory variables	Coefficient	Pr(> t )	95% CI
Cyanobacteria % total biomass	-75.9	0.58	Intercept	-1.725	0.049 *	-3.442, -0.008
			<b>avg. nitrate (<math>\text{NO}_3^-</math>, ug/L)</b>	<b>0.001</b>	<b>0.007 **</b>	<b>0.0004, 0.002</b>
			avg. phosphate ( $\text{PO}_4$ , ug/L)	-0.0003	0.197	-0.001, 0.0002
			avg. bicarbonate ( $\text{HCO}_3^-$ , ug/L)	-0.00001	0.079	-0.00002, 0.000001
			<b>water temperature (<math>^\circ\text{C}</math>)</b>	<b>0.072</b>	<b>0.007 **</b>	0.022, 0.123
Diatom % total biomass	-63.5	N/A	Intercept	0.319	<0.001 ***	0.202, 0.436
Dinoflagellate % total biomass	-69.1	0.25	Intercept	1.652	0.067	-0.126, 3.430
			<b>avg. nitrate (<math>\text{NO}_3^-</math>, ug/L)</b>	<b>-0.001</b>	<b>0.012 *</b>	<b>-0.002, -0.0003</b>
			water temperature ( $^\circ\text{C}$ )	-0.041	0.154	-0.098, 0.016

Table includes the AIC and  $R^2$  values, coefficients, and 95% confidence intervals of the best percent total biomass (measured as biovolume) models for three algal groups: cyanobacteria, diatoms, and dinoflagellates. The model was chosen using stepwise AIC model selection. Significant variables (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ ) are in bold



**Fig. 7** Relationship between nitrate concentration and relative cyanobacteria (left) and dinoflagellate (right) biomass (measured as biovolume). Points represent visits to each site ( $n = 26$ ); best fit lines represent the relationships between nitrate concentration and cyanobacteria and dinoflagellate relative bio-

mass, and gray shading represents a 95% confidence interval. Relative cyanobacteria biomass increased significantly with  $\text{NO}_3^-$  concentration ( $p < 0.01$ ) while percent biomass of dinoflagellates decreased ( $p < 0.05$ ). See Table 6 for slopes based on full model

### Nitrate and total phosphorus over time

The contrasting trends in nitrate and TP over time suggest that the two nutrients are influenced by different factors, like different sources or mobility. Nitrate concentrations decreased over time in the Chowan County sites, especially after Hurricane Isaias, indicating a dilution effect. Similar dilution trends were found with lower pesticide and nutrient levels, including TN and nitrate-N, in North Carolina's lower Cape Fear River watershed, where concentrated animal feeding operations (CAFOs) largely contribute to nutrient loading, after Hurricane Floyd (Mallin et al. 2002; Mallin and Corbett 2006). Mallin and others

(2002) suggest these low concentrations were due to denitrification (the conversion of  $\text{NO}_3^-$  to atmospheric  $\text{N}_2$ ; Burgin et al. 2011) in the floodplain. High turbidity from sediment disturbed during a storm limits light penetration into water and photosynthesis (Blain and Shears 2019; Schmidt et al. 2019), which can lead to hypoxic conditions that promote denitrification. After Hurricane Isaias, the water in the sound was cloudy and brown (Fig. S8), indicating mobilization of sediments. Thus, the decline in nitrate concentration across study sites observed in this study could also be due to denitrification.

Unlike nitrate, TP increased over time, especially after the hurricane. While heavy precipitation may

cause a dilution of environmental contaminants, it has also been found to increase pollutant levels in waterways depending on surrounding land use. Some sites in North Carolina demonstrated above-average TN, TP, and ammonia levels after Hurricanes Fran, Bonnie, and Floyd. These nutrients could largely be traced to concentrated animal feeding operations (CAFOs) and human sewage (Mallin and Corbett 2006). The lack of dilution effect suggests that greater rainfall caused mobilization of a major source of phosphorus on land. Erosion or resuspension of sediment is a potential explanation for this observed increase in TP (Carpenter et al. 1998; Cordell et al. 2009; Peñuelas et al. 2013; Ulén and Kalisky 2005). Fertilizer application can influence soil P concentrations (Carpenter et al. 1998), so P levels may be higher in agricultural soils. In investigating the potential effects of various agricultural and climate change scenarios on N and P loading into the Baltic Sea, Huttunen et al. (2015) found that soil erosion and erosivity played a role in TP loading from agricultural soils. In addition to increasing turbidity facilitating denitrification, resuspension of sediments after a storm can release P from soils (House et al. 1998; Stutter et al. 2008). These results indicate that sources of phosphorus and nitrogen in the sound differ.

### Land use and spatial variation

Nitrate was not significantly associated with the proportion of agricultural land use in a site's watershed. In this region, agricultural land is overwhelmingly cultivated cropland (NLCD2016, *n.d.*). According to county profiles from the 2017 Census of Agriculture, the mean proportion of cropland in the eight counties included in the study area was 89% (predominantly soy and grains), compared to 1% pastureland and 8.25% woodland (Census of Agriculture—State and County Profiles—North Carolina 2017). The proportion of agricultural land cover in study site watersheds ranged between 0.24 and 85.79%. The lack of relationship between nitrate concentration and land use was surprising considering the prevalence of industrial agriculture in the region and variation in watershed land use (Savci 2012). According to the best fit model, none of the variables included in this study explained variation in nitrate concentrations. While agricultural fertilizers are known to contain

nitrogenous compounds, the fertilizers used on farms in the study region are unknown. One fertilizer often used in the southeastern USA is a mix of urea and ammonium nitrate (called UAN; Environmental Defense Fund, *n.d.*). Determining which fertilizers are used on the farms surrounding the Albemarle Sound may help explain this result. Additionally, other factors may affect nitrate levels across the sound. Hobbie et al. (2017) found that atmospheric N deposition accounted for 34% of N inputs in seven urban watersheds, making it the second largest source of N. This atmospheric N comes from burning fossil fuels (Hobbie et al. 2017) and may be less relevant in more rural regions with lower population densities. However, there are other sources of nitrogen gases, like CAFOs and crop residue (Leytem et al. 2011; Tubiello et al. 2013). These potential sources of nitrate in the Albemarle region should be studied further.

Our results indicated that P increased with distance to river mouth (i.e., distance upstream). This finding contrasts with previous research that suggests higher phosphorus availability in coastal waters due to decreased phosphorus sequestration by iron (Blomqvist et al. 2004). For example, while Teichberg et al. (2010) found that P additions affected the growth of a macroalgal species in coastal waters, the researchers concluded that DIN was the main factor influencing growth, suggesting P was not limiting. In their meta-analysis, Elser et al. (2007) likewise observed that marine autotrophs are more limited by N than P, but that P-limitation may still occur in marine habitats. Because we do not know the TN concentrations at each site, it is difficult to determine the relative nutrient availability of these downstream sites, but the fact that ammonia followed the same pattern as phosphate suggests overall lower nutrient concentrations closer to the coast, potentially resulting in less algal growth than in upstream sites.

### Climate change, water quality, and algae

Our results demonstrate that impacts associated with climate change, specifically increasing frequency and intensity of extreme weather events and rising water temperatures significantly influence algal community composition in the Albemarle Sound.

First, the negative relationship between richness and precipitation points to a dilution effect, similar

to the apparent (but nonsignificant) influence of precipitation on  $\text{NO}_3^-$  concentration. Previous research in Danish lakes has demonstrated that precipitation can have a negative impact on diatom richness and lead to increased dominance of cyanobacteria (Özkan et al. 2013). This suggests that precipitation does not impact all algal taxa in the same way but can create conditions that favor the growth of certain taxa and lead to a decline in others, which would result in lower taxa richness.

The influence of precipitation on genus richness likely has implications for algal biomass. Decreased algal richness due to a dilution effect after heavy precipitation may create ideal conditions for one or few types of algae to dominate, leading to a bloom, especially if nutrient pollution from land enters the water as runoff (Özkan et al. 2013). While algal biomass in this study was not significantly influenced by precipitation, Lehman (2000) found that the biovolume of multiple algal taxa is greater with increased rainfall. Because the linear model only considered total algal biomass, changes in biomass of specific taxa in response to precipitation may not have been detected.

Total biomass was also not significantly related to nutrient concentration, yet *N* and *P* compounds are commonly known to stimulate algal growth (Altman and Paerl 2012; Bennett et al. 2001; Cordell et al. 2009; Galloway et al. 2003; Glibert et al. 2014; Savci 2012). The lack of relationship between biomass and nutrient concentration may be due to the inability to include TN and TP in the biovolume model. Only three *N* and *P* compounds were included in this analysis, so other nutrients may be influencing algal growth in this region. Additionally, TP was the only nutrient found to increase significantly with precipitation. Precipitation is a major pathway of nutrient loading into water, so TP is an important piece in analyzing this effect on algal biomass. Because extreme precipitation events are expected to increase in many regions including the southeastern USA due to climate change (Carter et al. 2018; Hayhoe et al. 2018), the influence of precipitation on algal community composition should be studied further.

Sites 5 and 6 had the lowest mean genus richness but the greatest mean biomass, demonstrating a potential inverse relationship between the two variables. Previous research has shown that phytoplankton richness is generally greatest at intermediate

biovolumes (Passy and Legendre 2006; Spatharis et al. 2008). Passy and Legendre (2006) suggest that phytoplankton richness is lower at high biomasses in both nutrient-rich and nutrient-poor waters because phytoplankton grow in environments with relatively few ecological niches. With fewer niches to occupy, there is more competition between species as biomass increases.

Total algal biomass and relative biomass of cyanobacteria increased significantly with water temperature. This finding aligns with previous research which demonstrates that algae thrive in warmer water compared to some aquatic plants (Arnold et al. 2017; Beer et al. 1986; Paerl and Otten 2013). This relationship between water temperature and algae could threaten aquatic trophic interactions reliant upon submerged aquatic vegetation (SAV). While aquatic macrophytes can be more competitive in nutrient-poor environments (Barko and James 1998; van Donk and van de Bund 2002), Beer et al. (1986) found that the filamentous algae *Lyngbya* favors warmer temperatures, with an optimal temperature of 40 °C compared to 30 °C for the macrophyte *Hydrilla*. Increased nutrient loading into water bodies from human activities and higher water temperatures due to climate change may optimize algal growth (Paerl and Otten 2013) and result in a decline in aquatic macrophytes.

Our results also indicate that water quality influences algal taxa differently. For instance, the relative biomass of cyanobacteria increased significantly with nitrate concentration while relative dinoflagellate biomass was negatively associated with nitrate. While cyanobacterial blooms are sometimes caused by N-fixers and therefore are expected to respond more to P availability (Schindler 1977), the growth of non-N-fixers like *Microcystis* has been shown to increase with the addition of *N* compounds like nitrate and ammonium (Rückert and Giani 2004). While all N-fixing genera in this study were cyanobacteria, the non-N-fixing genus *Microcystis* largely dominated the group. Therefore, the relative biomass of nitrogen-fixing cyanobacteria did not greatly influence how the phylum responded to nutrient concentrations.

The inverse response of cyanobacteria and dinoflagellates to  $\text{NO}_3^-$  concentration may be because an increase in the proportion total biomass of cyanobacteria would cause a decrease in the proportion of dinoflagellate biomass and vice versa. Dinoflagellates and cyanobacteria seem to be

particularly inversely related in sites 5 and 6, where cyanobacteria composed a large proportion of the total biomass in late July and were far less prevalent in early August when the relative biomass of dinoflagellates increased. This decline in relative cyanobacteria biomass during this time may be due to a dilution effect after Hurricane Isaias (Phlips et al. 2007), which similarly impacted nitrate concentration. Özkan et al. (2013) found the opposite effect of precipitation on cyanobacteria dominance in freshwater Danish lakes, but in our sites, saltwater intrusion following the hurricane may have created conditions favoring dinoflagellate genera, which most often thrive in marine ecosystems (de Vernal et al. 2013; White and Kaplan 2017). While conductivity was actually lower at sites 5 and 6 in August compared to other weeks, sampling was conducted several days after the hurricane, so these data may not have captured the impact of saltwater intrusion at those sites. More research in this region should examine the influence of conductivity and salinity on algal community composition.

Water quality and algal richness and biomass models were run excluding the last samples taken from each Chowan County site after Hurricane Isaias to illuminate the effects of the hurricane on water quality and better understand trends in non-hurricane conditions. TP concentration was significantly positively associated with proportion of agricultural land cover in the watershed, but not proportion of developed land cover (Table S9). The relationship between developed land and water quality may be stronger with the inclusion of post-hurricane data due to greater stormwater runoff into the sound during the hurricane. This change suggests that agricultural effects on water quality in this region are dominant until a major precipitation event. Additionally, algal richness was significantly negatively associated with conductivity (Table S10), while total algal biomass increased significantly with conductivity (Table S11). These relationships were not significant with post-hurricane data included. This difference may be because there was lower conductivity at each of the Chowan sites after the hurricane, which may have obscured these relationships. The positive association between biomass and conductivity may indicate a higher occurrence of algal blooms at sites closer to the coast or that high salinity gives few salt-tolerant taxa a competitive advantage over other algae. The specific taxa

we analyzed (cyanobacteria, diatoms, and dinoflagellates) did not exhibit any significant relationship with conductivity (Table S12), so it is unclear which taxa are driving this relationship. More information on the hurricane exclusion analyses can be found in the supplementary information file.

### Management implications

Our data indicate that climate change is a critical factor influencing water quality and algal community composition. Local management solutions alone are not enough to mitigate eutrophication and HABs.

The positive relationship between TP and precipitation indicates that erosion reduction could help reduce nutrient loading in the Albemarle Sound. Soil erosion can carry both organic and inorganic *P* compounds to waterbodies during precipitation events (Howarth et al. 2002; Huttunen et al. 2015). There are multiple methods of reducing erosion that have been employed in coastal ecosystems, like bulkheads, seawalls, and “living shorelines” (Beuzen et al. 2018; Bilkovic et al. 2016; Jackson et al. 2015; Scyphers et al. 2011). Furthermore, previous studies have shown that erosion mitigation measures may be related to water quality improvement (Hunt et al. 2019; Rickson 2014). Our data suggest that the link between erosion reduction and nutrient pollution is worth exploring in the Albemarle Sound.

TP was positively associated with developed and agricultural land use, indicating a potential link between farm and lawn fertilizers, impervious surfaces, and/or yard and pet waste. To reduce phosphorus inputs into the sound, individuals can dispose of their pet and yard waste, rather than leaving it on their lawn or street, where nutrients can leach into the water (Hobbie et al. 2017). Additionally, conserving and expanding greenspace and limiting impervious surface area may help precipitation to percolate into the soil over a greater area, which can reduce runoff (Liu et al. 2014). Increasing green infrastructure, like reservoirs and ponds, has also been shown to effectively limit storm runoff (Liu et al. 2014). Because conventional agricultural methods greatly contribute to nutrient loading (Chakraborty et al. 2017; Glibert et al. 2014), implementing farming methods that use fewer chemical fertilizers and farming practices that reduce nutrient export like cover crops and two-stage ditches, can be beneficial in reducing nutrient inputs

and eutrophication (Abdalla et al. 2019; Christopher et al. 2017; Reganold and Wachter 2016).

Protecting SAV can also be a useful method in algal bloom mitigation as macrophytes can serve as nutrient sinks in aquatic environments (Ozimek et al. 1990, Touchette and Burkholder 2000, van Donk et al. 1993). In cases where sources of nutrients on land are unclear, methods that reduce nutrient concentrations from within water bodies may be helpful. For instance, as our data did not indicate a clear source of nitrogen to the sound, protecting SAV could be useful in mitigating cyanobacterial blooms (particularly non-N-fixing genera). Some boat ramps in the study region included informational signs urging visitors to take individual actions to protect aquatic grasses, like avoiding shallow areas in one's boat (Fig. S9). We suggest implementing more signage like this to educate the public about protecting the waterways they use.

#### Research limitations and future directions

We faced multiple challenges and limitations in conducting this study. One, sites were selected based on accessibility, which limited the options available. Samples closer to the shore (e.g., sites 1, 2, 9, 20) may be more greatly influenced by waters from upstream rather than land use in their watersheds. Thus, further investigation into how the factors influencing water quality may vary based on a site's position in the landscape is warranted. Furthermore, the Albemarle region is limited in terms of land use composition. There are not many cities in the study area, and therefore it is difficult to determine the impact of urban development on water quality. The Chesapeake Bay is a comparable estuarine system surrounded by more densely populated cities. There is evidence that the effects of urbanization on water quality in this watershed depend on land use history (e.g., is urban development expanding into forests or agricultural land; Ator et al. 2020). Studying the effects of urban land area in the Chesapeake Bay would improve our understanding of how urbanization affects water quality in brackish water estuaries. We analyzed preserved algal samples rather than fresh samples, which may have impacted the morphology of some algal taxa and complicated taxa identification. Lastly, we were not able to obtain TN data due to COVID-19 restrictions,

which limits our knowledge of how *N* concentration relates to land use and algal community composition.

While understanding the factors that impact the concentrations of various nutrients and algae in the sound provides crucial insight into managing algal blooms, more research should be conducted to better understand the influence of space, time, and land use on algal blooms. Exploring N:P ratios in the sound would be useful in understanding variation in algal composition and growth, especially for cyanobacteria. Sampling runoff directly and sampling at different depths would also provide more information regarding which nutrients and pollutants are entering the sound and their impact on water quality. Furthermore, the variables analyzed in this study did not appear to best explain spatial variation in  $\text{NO}_3^-$  concentrations. However, the model of the association between nitrate and proportion agricultural land fits the data just as well as the best model (Table S2; Burnham and Anderson 2002). Future research should examine this relationship further and focus on how other factors, like conductivity, temperature, pH, and even air quality affect nitrate. Lastly, analyzing water quality and algal community composition data in the sound over longer periods of time will help in determining the relationship between climate change, land use, water quality, and algal growth.

#### Conclusion

Nutrient concentrations in the Albemarle Sound are impacted by varying factors. Nitrate, ammonia, and phosphorus were not significantly associated with the same variables, meaning they may come from different sources. Agricultural fertilizers commonly contain inorganic forms of nitrogen and phosphorus, but nitrate was not significantly associated with proportion of agricultural land use and declined over time in the six sites monitored biweekly even after a large precipitation event, signifying a dilution effect that was not offset from atmospheric *N* inputs. Total phosphorus showed an opposite relationship with time and increased with proportion farmland. The increase in TP with precipitation may be in part due to erosion. In general, these results illustrate a need for further research into sources of nutrient pollution in the sound.

Algal community composition was influenced by nitrate concentration, water temperature, and precipitation, suggesting that climate change plays an important role in algal growth. Hurricane Isaias demonstrated the impact extreme weather events, leading to a dilution effect with algal richness and potentially influencing the relative biomass of algal taxa. Total biomass increased with water temperature, demonstrating that current warming trends will likely support excessive algal growth at the expense of other aquatic organisms.

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