MANAGEMENT APPLICATIONS



Using Spatial Variability in the Rate of Change of Chlorophyll *a* to Improve Water Quality Management in a Subtropical Oligotrophic Estuary

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Abstract

Anthropogenic eutrophication threatens numerous aquatic ecosystems across the globe. Proactive management that prevents a system from becoming eutrophied is more effective and cheaper than restoring a eutrophic system, but detecting early warning signs and problematic nutrient sources in a relatively healthy system can be difficult. The goal of this study was to investigate if rates of change in chlorophyll *a* and nutrient concentrations at individual stations can be used to identify specific areas that need to be targeted for management. Biscayne Bay is a coastal embayment in southeast Florida with primarily adequate water quality that has experienced rapid human population growth over the last century. Water quality data collected at 48 stations throughout Biscayne Bay over a 20-year period (1995–2014) were examined to identify any water quality trends associated with eutrophication. Chlorophyll *a* and phosphate concentrations have increased throughout Biscayne Bay, which is a primary indicator of eutrophication. Moreover, chlorophyll *a* concentrations throughout the northern area, where circulation is restricted, and in nearshore areas of central Biscayne Bay are increasing at a higher rate compared to the rest of the Bay. This suggests increases in chlorophyll *a* are due to local nutrient sources from the watershed. These areas are also where recent seagrass die-offs have occurred, suggesting an urgent need for management intervention. This is in contrast with the state of Florida listing of Biscayne Bay as a medium priority impaired body of water.

Keywords Chlorophyll a · Eutrophication · Oligotrophic · Ecological indicators

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Introduction

Eutrophication, excessive growth of plants and algae caused by the enrichment of nutrient concentrations in aquatic systems, is a well-documented phenomenon that affects a range of aquatic systems (Bricker et al. 1999; Kemp et al. 2005; Diaz and Rosenberg 2008). Eutrophication has been linked to increased frequency of low dissolved oxygen, harmful algae blooms, and seagrass die-off (Hagy et al. 2004; Kemp et al. 2005; Li et al. 2015). If increases in nutrient concentrations continue, eventually, a regime shift may occur where pelagic primary production dominates and a system can settle into a new stable state (Zhang et al. 2003). Once a regime shift occurs, restoration efforts can be expensive and it is possible a system will not fully recover (Duarte et al. 2009). There is also greater policy efficiency and more political and economic benefits by avoiding regime shifts (Kelly et al. 2014). To avoid a eutrophication regime shift, major sources of nutrients must be identified and a management plan implemented as soon as possible.



Chlorophyll a concentration is an accepted indicator of eutrophication that can be examined to assess if the input of anthropogenic nutrients is affecting an ecosystem. Chlorophyll a is a primary indicator; it responds to increasing inputs of nutrients before more serious and difficult to reverse damage has occurred, such as loss of seagrass and other submerged aquatic vegetation (Boyer et al. 2009). In oligotrophic systems, or systems with low nutrient and chlorophyll a concentrations, inputs of nutrients are typically taken up immediately by the benthic community, biofilms, or phytoplankton, making an increase in dissolved nutrients undetectable. Thus, chlorophyll a, a direct proxy for phytoplankton biomass, is typically used to indicate the trophic status of a coastal ecosystem (Steele 1962; Cullen 1982; Boyer et al. 2009). Elevated phytoplankton biomass is one of the most visible and early symptoms of eutrophication, and long-term increasing trends of phytoplankton biomass (chlorophyll a) can be an indication that nutrient inputs into a system are increasing (Bricker et al. 1999; Fisher et al. 2006; Boyer et al. 2009).

Biscayne Bay is a shallow, oligotrophic bay in southeastern Florida adjacent to the city of Miami and along the coast of the Atlantic Ocean (Fig. 1). Between 1994 and 2003, the median chlorophyll *a* concentration within Biscayne Bay was reported to be 0.28 μg L⁻¹ (Caccia and Boyer 2005). Unlike most estuaries, Biscayne Bay phytoplankton growth is limited primarily by phosphorous availability (Brand et al. 1991; Kleppel 1996; Carey et al. 2011). P-limitation of Biscayne Bay has been found based on long-term monitoring (Caccia and Boyer 2007) and from nutrient bioassays throughout Biscayne Bay (Brand et al. 1991).

Over the twentieth century, the Biscayne Bay watershed has experienced rapid growth in its human population, which resulted in increased urbanization, agricultural activities, and changes in water management practices (SFWMD 1995). Although earlier studies (Bricker et al. 1999; Caccia and Boyer 2005) indicated relatively satisfactory water quality conditions, more recent data indicates decreases in water quality. Average chlorophyll a and inorganic nutrient concentrations are lower compared to other systems with watersheds altered by anthropogenic activity (Caccia and Boyer 2005; Cloern and Jassby 2010; Harding et al. 2016). However, the state of Florida recently recognized that chlorophyll a levels in Biscayne Bay exceed the state water quality criterion, triggering the need for restoration by developing a total maximum daily load (TMDL) (Florida Department of Environmental Protection 2018). Further, there are localized indications that poor water quality is affecting the Biscayne Bay ecosystem, such as periods of low oxygen and loss of seagrass species associated with elevated nitrogen concentrations in specific areas of South Biscayne Bay (Meeder and Boyer 2001). Another indication is a die-off of seagrass in North Biscayne Bay (Avila et al. 2017), a relatively enclosed region of Biscayne Bay known for high chlorophyll a concentrations

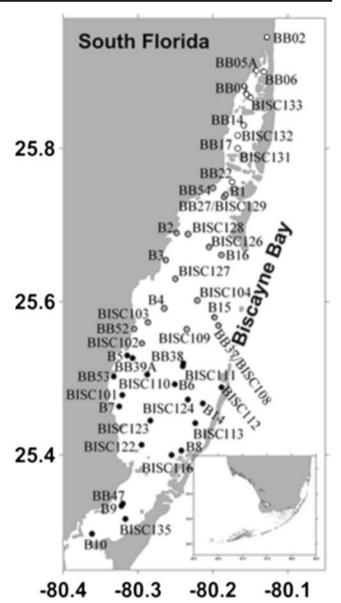


Fig. 1 Location of 48 water quality stations in Biscayne Bay

compared to the rest of the Bay (Caccia and Boyer 2005). There has also been a, persistent macroalgal bloom of *Anadyomene* J.V. Lamouroux that left historic seagrass areas unvegetated (Collado-Vides et al. 2013). Biscayne Bay has numerous potential sources of nutrients, and it is unknown which specific nutrient sources are affecting water quality, or if they are affecting water quality at all. This makes Biscayne Bay an ideal system to assess if spatial variability in trends of chlorophyll *a* is an effective management tool.

The identification of a long-term, systematic increase in average chlorophyll *a* concentrations could be a primary indication that Biscayne Bay is becoming more eutrophic, which could eventually result in an ecosystem regime shift (McQuatters-Gollop et al. 2007). A secondary indication that Biscayne Bay could be headed towards a regime shift would



be a loss of seagrass, as decrease in water quality shifts the system from a benthic-dominated to a pelagic-dominated system (Zhang et al. 2003; Barbier et al. 2011). The loss of seagrasses creates a positive feedback loop whereby seagrass mortality results in increased re-suspension of nutrients and sediments further shading seagrasses and providing nutrients to perpetuate the phytoplankton bloom (Glibert et al. 2009; Millette et al. 2018). As mentioned earlier, major losses of seagrass have recently occurred in specific regions of Biscayne Bay (Avila et al. 2017), so it would be useful to consider if any of the changes in chlorophyll *a* or nutrient concentrations that we identify are co-located with these loss of seagrasses.

We assessed chlorophyll a and nutrient data collected by three different agencies at forty-eight stations (Fig. 1) throughout Biscayne Bay between 1995 and 2014 to examine longterm trends in chlorophyll a and nutrient concentrations. This analysis was used to address two specific questions: (1) How widespread are early indications of eutrophication and which, if any, areas are more affected by eutrophication compared to others? (2) How does a traditional regional-scale analysis of long-term trends compare to analysis of individual stations within that region? Then, we used our analysis from these questions to assess how effectively the current method used by the state of Florida to identify impaired bodies of water classifies the status of Biscayne Bay. It is common practice, and legally mandated in the state of Florida, to group data from water quality stations in a similar region for analysis (Caccia and Boyer 2007; Brandt et al. 2014; Harding et al. 2015, 2016; Fla. Admin. Code 2016). By analyzing long-term trends at individual stations, we were able to identify if and where water quality was most degraded and assess how our description of the status of water quality in Biscayne Bay compares to the state of Florida's description.

Methods

Site Description Biscayne Bay has historically been divided into three regions based upon similarities in environmental conditions: North Biscayne Bay (NBB), Central Biscayne Bay (CBB), and South Biscayne Bay (SBB) (Caccia and Boyer 2005). The dense urban core of the Miami area is located on the coast of NBB, making the watershed for this region of Biscayne Bay the most urbanized (DERM 1981). CBB is considered a transition zone from NBB to SBB. The watershed surrounding this region is suburban and urban, but less dense than NBB (Caccia and Boyer 2005). The SBB watershed is predominantly agricultural (Caccia and Boyer 2005). There is a strong north-south gradient of chlorophyll *a* from NBB to SBB, with chlorophyll *a* concentrations three times higher in NBB (Caccia and Boyer 2005). In the CBB and SBB, there is an onshore-offshore gradient of chlorophyll

a concentrations, with concentrations higher nearshore (Caccia and Boyer 2005).

Observations There are ninety-two water quality stations throughout Biscayne Bay that have been sampled with varying degrees of consistency by three different agencies, the National Oceanic and Atmospheric Administration (NOAA), the Miami-Dade County Department of Environmental Resources Management (DERM), and the Southeast Environmental Research Center of Florida International University (SERC FIU), in conjunction with the South Florida Water Management District (SFWMD). In this study, the data records from forty-eight of the more consistently sampled stations (approximately once a month) collected between 1995 and 2014 were analyzed. This time period was selected to examine long-term trends because it occurs within a stable positive phase of the Atlantic Multidecadal Oscillation (AMO), which affects chlorophyll a in the adjacent embayment of Florida Bay (Briceño and Boyer 2009). A drastic decrease in chlorophyll a concentrations during 1994 was attributed to a shift in the AMO phase from negative to positive (Briceño and Boyer 2009). We selected forty-eight stations to analyze based on the availability of sufficient chlorophyll a data during the 20-year study period: ten stations in NBB, eighteen stations in CBB, and twenty-one stations in SBB (Table 1; Fig. 1).

Stations were deemed to have sufficient data if they had > 10 years of data that had been collected for 10 continuous years at any point between 1995 and 2014. The parameters collected and methods used for collection at each station varied based on which agency was sampling the station (Table 2). Analyses were conducted on parameters that could be affected by anthropogenic nutrient inputs and were collected consistently at most stations with no apparent bias between different methods: chlorophyll a, nitrate + nitrite (NO_x), ammonium (NH₄⁺), and soluble reactive phosphate (PO₄³⁻) concentrations. We used NO_x, NH₄⁺, and PO₄³⁻ concentrations collected at each sampling data to calculate a DIN:DIP ratio. All stations we analyzed sampled chlorophyll a concentrations, but only thirty-seven stations collected samples for nutrients (Table 1). Of the thirty-seven nutrient stations, thirteen stations had too many gaps in the time-series for reliable long-term analysis. These thirteen stations were included in spatial analysis of concentrations, but were excluded from the long-term analysis.

Ten of the water quality stations were sampled by SERC FIU between January 1995 or June 1996 and September 2009. Starting in January 2010, the stations were sampled by SFWMD (Table 1). The sampling and analysis methods remained the same; however, visual inspection of the data showed a shift in the pattern of nutrient data at all stations at the same time the change in



Table 1 The list of stations in Biscayne Bay we used in our analysis, the agency in charge of collecting and analyzing data at each station, the range of years for which chlorophyll a data was available, the number

of data points throughout the time series, and the number of missing data points throughout the time series

Station name	Region	Collection agency	Years of data	No. of data points	No. of missing data points
BB02	NBB	DERM	1995–2014	219	21
BB05A	NBB	DERM	1997-2014	196	20
BISC134	NBB	SERC FIU/SFWMD	1996-2014	214	6
BB09	NBB	DERM	1995-2014	220	20
BISC133	NBB	SERC FIU/SFWMD	1996-2014	210	10
BB14	NBB	DERM	1995-2014	217	15
BISC132	NBB	SERC FIU	1996-2007	133	3
BB17	NBB	DERM	1995-2014	220	20
BISC131	NBB	SERC FIU/SFWMD	1996-2014	209	11
BB22	NBB	SERC FIU/SFWMD	1996-2014	217	6
BB54	CBB	DERM	2004-2014	119	2
B1	CBB	NOAA/AOML	2002-2014	75	68
BISC129	CBB	SERC FIU/SFWMD	1996-2014	212	8
B2	CBB	NOAA/AOML	2002-2014	73	69
BISC128	CBB	SERC FIU	1996–2009	152	8
BISC126	CBB	SERC FIU	1996–2007	131	5
B16	CBB	NOAA/AOML	2002–2014	73	70
B3	CBB	NOAA/AOML	2002–2014	75	67
BISC127	CBB	SERC FIU	1996–2009	151	9
BISC104	CBB	SERC FIU	1995–2008	160	5
B4	CBB	NOAA/AOML	2002–2014	72	70
B15	CBB	NOAA/AOML	2002–2014	73	70
BISC103	CBB	SERC FIU	1995–2007	148	5
BISC108	CBB	SERC FIU/SFWMD	1995–2014	231	9
BB52	CBB	DERM	1999–2014	178	11
BISC109	CBB	FIU	1995–2007	148	5
BISC102	CBB	SERC FIU/SFWMD	1995–2014	220	17
B5	SBB	NOAA/AOML	2002–2014	75	66
BB39A	SBB	DERM	1997–2014	195	21
BB39A BB38	SBB	DERM	1995–2014	220	20
BISC111	SBB	SERC FIU/SFWMD	1995–2014	223	15
BISC111	SBB	SERC FIU/SFWMD	1995–2014	221	17
BB53	SBB	DERM	1999–2014	173	16
B6	SBB	NOAA/AOML	2002–2014	72	70
BISC112	SBB		1995–2008	160	5
BISC112 BISC101	SBB	SERC FIU SERC FIU/SFWMD		230	10
BISC101 BISC124	SBB	SERC FIU	1995–2014 1995–2007	148	5
B14	SBB	NOAA/AOML	2002–2014	74	68
B7	SBB	NOAA/AOML	2002–2014	75	68
BISC123	SBB	SERC FIU/SFWMD	1995–2014	226	12
BISC113	SBB	SERC FIU	1995–2007	148	5
BISC122	SBB	SERC FIU/SFWMD	1995–2014	227	13
B8	SBB	NOAA/AOML	2002–2014	75	68
BISC116	SBB	SERC FIU	1995–2007	148	5
BB47	SBB	DERM	1995–2014	220	20
B9	SBB	NOAA/AOML	2002–2014	73	70
BISC135	SBB	SERC FIU	1996–2009	153	8
B10	SBB	NOAA/AOML	2002–2014	75	68



Table 2 A list of the instruments and laboratory methods used to collect and analyze the data collected by AOML, SERC FIU/SFWMD, and DERM and used in this paper. DERM did not report any nutrient data;

therefore, the method used to analyze nutrient concentrations is not listed. For chlorophyll *a*, NO_x, NH₄⁺, and PO₄³⁻ concentrations, the methods used to analyze the samples in the laboratory are reported

	Chl a	NO _x	NH ₄ ⁺	PO ₄ ³⁻
AOML	Filtration extraction using a 60:40 mixture of acetone and dimethyl sulfide (Shoaf and Lium 1976)	EPA Methods 353.4	EPA Methods 349.0	EPA Methods 365.5
SERC FIU/SFWMD	Filtration extraction using a 90% acetone (Strickland and Parson 1972)	Alpkem model RFA 300 (Caccia and Boyer 2005, 2007)	Alpkem model RFA 300 (Caccia and Boyer 2005, 2007)	Alpkem model RFA 300 (Caccia and Boyer 2005, 2007)
DERM	Filtration extraction using a 90% acetone (EPA Methods 445.0)	N/A	N/A	N/A

sampling agency occurred (Fig. 2). There was no noticeable shift in chlorophyll *a* concentrations (Fig. 2). Because the cause of the shift in the pattern of data is unknown and could be the result of the change in the agency doing the collection and analysis, we decided to only analyze nutrient data from before 2010. This means that analysis of long-term trends for nutrients at any of the stations is only for the time period of 1995–2009.

Treatment of Data For the initial analysis, we averaged data across each month for all stations in each region of Biscayne Bay (NBB, CBB, and SBB). Next, we analyzed each station individually in order to identify small-scale spatial variability in long-term trends of water quality parameters.

Statistical Analysis We examined if water quality parameters in Biscayne Bay showed any notable temporal trends using a

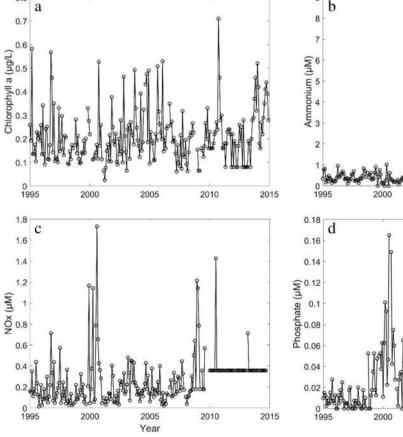
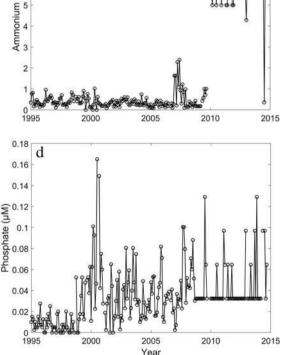


Fig. 2 The time series, with outliers removed[#], for a chlorophyll a concentrations (μ g L⁻¹), **b** NO_x (μ M), **c** NH₄⁺ (μ M), and **d** PO₄³⁻ (μ M) at station BISC108. BISC108 is one of the ten stations that were sampled by SERC FIU through 2009, and then in 2010, SFWMD took over sampling. While the sampling and analysis methods remained the



same after the switch, there is still a visible change in all nutrient concentrations most likely associated with the change in the sampling agency. **Outliers were removed from each dataset in order to visually highlight how the shift in sampling agencies affected the nutrient data. However, no data was removed for any statistical analysis



seasonal Mann-Kendall (SMK) test. The SMK is a nonparametric test that is a recommended method for identifying trends in water quality datasets with strong seasonality (Meals et al. 2011). We used the "kendallSeasonalTrendTest" from the R package, EnvStats (Millard 2017), to calculate the Theil-Sen slope for each factor at every station. The Theil-Sen slope reflects the annual rate of change of a parameter over the period of time that data were collected. The "kendallSeasonalTrendTest" also calculated the 95% confidence intervals and p values for each Theil-Sen slope (Millard 2017). We report all of the slopes along with their 95% confidence intervals and p values in place of labeling a slope as significant or not significant. This was done because waiting for a slope to be considered significant based on an arbitrary criterion can increase management response time to a system that is likely experiencing significant shifts in water quality (e.g., the Precautionary Principle, Raffensperger and Tickner 1999).

Results

Regional Comparison There were statistically significant regional differences for all mean water quality parameters except mean phosphate and DIN:DIP (Fig. 3). Chlorophyll *a* had the largest regional differences with mean chlorophyll *a* concentration three times higher in NBB compared to CBB and SBB (Fig. 3). In contrast, mean NO_x concentrations were higher in SBB compared to CBB and NBB and mean NH₄⁺ was highest in NBB and lowest in CBB (Fig. 3).

Chlorophyll a concentrations increased in NBB (0.029 µg chl a L $^{-1}$ year $^{-1}$, LCI = 0.15, UCI = 0.42, Fig. 4a), CBB (0.023 µg chl a L $^{-1}$ year $^{-1}$, LCI = 0.18, UCI = 0.27, Fig. 4b), and SBB (0.013 µg chl a L $^{-1}$ year $^{-1}$, LCI = 0.11, UCI = 0.15, Fig. 4c). Based on the 95% confidence intervals, the rates of change were not different between NBB and CBB, but the rate of change in SBB was lower compared to NBB and CBB. PO $_4$ concentrations and DIN:DIP had rates of change between 1995 and 2009 in all regions with p values < 0.001, while NH $_4$ concentrations had rates of change with p values > 0.1 in all regions (Table 3). NO $_x$ concentrations had rates of change with p values < 0.05 and < 0.1 in SBB and CBB, respectively (Table 3). PO $_4$ concentrations were increasing in all regions and DIN:DIP was decreasing in all regions (Table 3).

Spatial Variability When individual station data were examined, mean chlorophyll a, NO_x , NH_4^+ , and PO_4^{3-} concentrations all followed the same general distribution pattern with the highest concentrations nearshore and decreasing concentrations towards offshore stations (Fig. 5). Here we define nearshore stations as the stations closest to land on the western side of the Bay. Chlorophyll a concentrations were highest in NBB (Fig. 5a). NO_x concentrations were highest in SBB (Fig. 5b), near stations

B5 (13.7 μ M), B7 (10.4 μ M), and BISC101 (9.5 μ M). NH₄⁺ concentrations were highest in SBB (Fig. 5c), near station B5 (8.53 μ M), B7 (4.14 μ M), and BISC101 (3.09 μ M). PO₄³⁻ concentrations were highest in CBB and SBB (Fig. 5d), near stations B2 (0.09 μ M), B3 (0.09 μ M), and B5 (0.11 μ M).

We calculated the annual rate of change in chlorophyll a, PO₄³⁻, NH₄⁺, and NO_x concentrations at all stations with sufficient data. For chlorophyll a concentrations, thirty-three stations had trends with a p value < 0.05 (all positive), 5 stations had trends with a p value < 0.1 (all positive), and 10 stations had trends with a p value > 0.1 (6 positive, 4 negative) (Table 4). The highest rate of increase, $0.046 \mu g chl a L^{-1} vear^{-1} (LCI =$ 0.23, UCI = 0.66), was at station BISC133 (Fig. 6a). The annual rate of change for chlorophyll a was high throughout most of NBB compared to CBB and SBB (Fig. 6a). Based on a comparison of the 95% confidence intervals, all but one of the rates of change at individual stations in NBB were similar to the regional rate of change (Table 4). In CBB and SBB, the annual rate of change of chlorophyll a was highest nearshore and decreased in the offshore direction, with the lowest annual rate of change where the Atlantic Ocean exchanges with Biscayne Bay (Fig. 6a). In CBB and SBB, chlorophyll a's annual rate of change was higher at nearshore stations B1, B2, B3, and B10 (Fig. 6a) compared to other stations. Twenty of the thirty-eight confidence intervals for the rates of change at individual stations in CBB and SBB did not overlap with the regional rate of change confidence interval (Table 4).

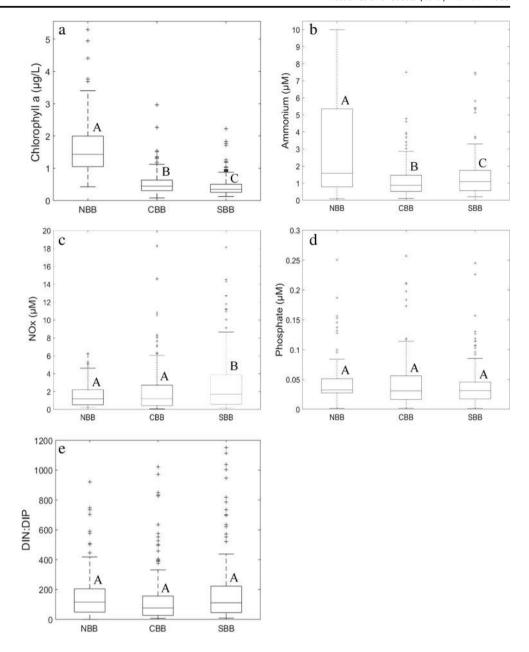
 NO_x concentrations had trends with p values > 0.1 at eighteen stations (10 positive, 8 negative), trends with p values < 0.1 at two stations (1 positive, 1 negative), and trends with p values < 0.05 at four stations (3 positive, 1 negative) (Fig. 6b; SI Table 5). The highest rates of increase occurred at nearshore stations BISC102 and BISC103 (Fig. 6b). NH₄⁺ annual rates of change were similar to NO_x rates of change (Fig. 6c). NH_4^+ concentrations had trends with p values > 0.1 at fifteen stations (6 positive, 9 negative), trends with pvalues < 0.1 at three stations (1 positive, 2 negative), and trends with p values < 0.05 at six stations (3 positive, 3 negative) (Fig. 6c; SI Table 6). The highest rates of increase occur at nearshore stations BISC101, BISC102, and BISC103 (Fig. 6c). In contrast, PO_4^{3-} concentrations had trends with p values < 0.05 at all twenty-four stations and they were all positive (Fig. 6d; SI Table 7). The rate of change in PO₄³⁻ was similar throughout Biscayne Bay, but the highest rate of increase in PO₄³⁻ concentrations occurred at stations BISC132 and BISC113 (Fig. 6d).

Discussion

The analysis of long-term trends at individual stations exposed a large degree of spatial variability that was not observed in the regional analysis, particularly for chlorophyll *a* concentrations.



Fig. 3 Box and whisker plots showing the mean, quartiles, and range of a chlorophyll a concentrations ($\mu g L^{-1}$), **b** NO_x (μM) , c NH_4^+ (μM) , d PO_4^3 (μ M), and e NO_x:PO₄³⁻ in the three distinct regions of Biscayne Bay: north Biscayne Bay (NBB), central Biscayne Bay (CBB), and south Biscayne Bay (SBB). Onefactor ANOVA tests were run for each factor to compare the different regions. The letters represent regions that are significantly different (p < 0.05) from each other for each parameter

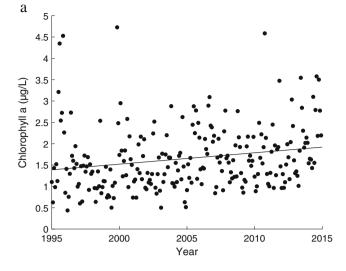


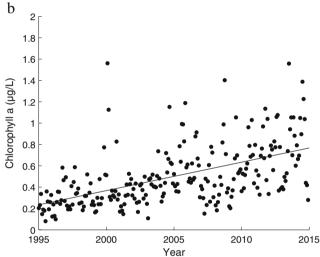
Over a 20-year period, from 1995 to 2014, chlorophyll a concentrations increased significantly in all three regions of Biscayne Bay. However, the rate of increase had high spatial variability with the largest increase occurring in NBB and at nearshore stations close to specific canal outflows in CBB. Through our analysis of chlorophyll a rates of change at individual stations, we were able to determine where water quality was deteriorating at the highest rate in Biscayne Bay. The location of the rates of increase in chlorophyll a concentrations likely indicates that local sources of nutrients are driving these long-term increases in chlorophyll a concentrations.

Previous research on water quality in Biscayne Bay assessed regional and seasonal variability in water quality by focusing on chlorophyll *a* and nutrient concentrations (Caccia and Boyer

2005; Boyer et al. 2009). The results novel to this study are the analysis of long-term trends at individual stations in Biscayne Bay. Earlier analysis of water quality showed that chlorophyll *a* concentrations are highest in NBB and along the shoreline in CBB and SBB (Caccia and Boyer 2005). Our analysis confirmed these findings, but also found that these are the same areas where chlorophyll *a* is increasing at the highest rate (Figs. 5 and 6). Additionally, our analysis of trends in nutrient concentrations demonstrated that long-term changes in inorganic nitrogen are not similar to changes in phosphate. Inorganic nitrogen had a wide range of rates of change between 1995 and 2009; however, there was a localized area consisting of four stations with noticeably higher rates of change compared to the rest of Biscayne Bay (Fig. 6). In contrast,







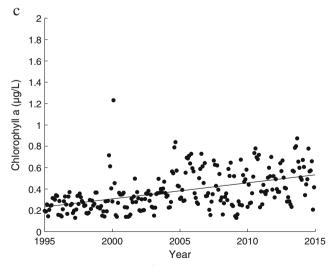


Fig. 4 The chlorophyll a (µg L⁻¹) concentrations in **a** NBB, **b** CBB, and **c** SBB from 1996 to 2014 in NBB and 1995 to 2014 in CBB and SBB

phosphate concentrations uniformly increased throughout all of Biscayne Bay during this same period (Fig. 6).

Analysis of long-term trends between a regional analysis and individual stations produced different results and conclusions. The regional analyses of long-term trends in chlorophyll a suggest that the annual rate of change was similar in NBB and CBB and lower in SBB (Fig. 4). However, the analysis of individual stations showed that CBB and SBB had high rates of increase nearshore with rapid decreases in the rate of change farther from land, such that most areas of SBB and CBB showed only slight increases (Fig. 6, Table 4). The rates of change in chlorophyll a at individual stations were more uniformly high in NBB compared to CBB and SBB (Table 4). The regional rate of change for NO_x and NH₄⁺ suggested that NH₄⁺ was likely not changing within the whole system, but that NOx likely increased in CBB and SBB (Table 3). However, analysis of individual stations showed more nuanced results for NO_x and NH₄⁺; both inorganic nitrogen species had high rates of change in the same CBB and SBB region (Fig. 6; SI Tables 5 and 6).

While water quality throughout most of CBB is not deteriorating as quickly when compared to NBB, there are two stations with high increases in chlorophyll a: B2 and B3. Both stations are located close to the mouth of canals that empty into Biscayne Bay, Coral Gables Waterway (B2) and Snapper Creek Canal (B3). This suggests that high concentrations of chlorophyll a and/or nutrients are entering into Biscayne Bay through these canals and these areas are becoming more eutrophic. It is possible that these stations are the beginning of major degradation in water quality that could spread to the rest of Biscayne Bay. There was a recent seagrass die-off event that occurred near stations B2 and B3 in 2005-2008 and seagrass is still struggling to recover (Collado-Vides et al. 2013), a potential secondary indication that this specific area could be headed towards a regime shift (Zhang et al. 2003). Identifying and managing the major sources of nutrients into Coral Gables Waterway and Snapper Creek Canal would be the obvious starting point to restoring water quality near stations B2 and B3.

The discrepancy between the two approaches supports an individual station approach to water quality analysis compared to a regional one because it provides more information and thus a more nuanced view of long-term trends in water quality. Previous analysis on water quality in Biscayne Bay has either grouped stations into three (Boyer et al. 2009), five (Caccia and Boyer 2005), or nine regions (Briceño et al. 2013). With a regional analysis, we still would have rightly concluded that chlorophyll *a* and phosphate concentrations are increasing, but our analysis of long-term trends at individual stations provide a detailed look at where water quality is degrading at the fastest rate.

Currently, the state of Florida has an established methodology, based on representative data, to identify impaired surface waters that will be included on the state's Verified List of impaired waters and have Total Maximum Daily Loads



Table 3 The annual rate of change (slope), lower and upper confidence intervals (LCI and UCI, respectively), and p value from 1995 to 2009 for phosphate (PO₄³⁻, μ M year⁻¹), nitrate + nitrite (NO_x, μ M year⁻¹), ammonium (NH₄⁺ μ M year⁻¹), and NO_x:PO₄³⁻ in the three different

regions of Biscayne Bay. Note that the annual rate for nutrients in NBB was calculated between 1996 and 2009 while the rate of change in CBB and SBB was calculated between 1995 and 2009. All values were estimated using the Theil-Sen slope from the seasonal Mann-Kendall test

	PO ₄ ³⁻			NO _x			NH ₄ ⁺			DIN:DIP						
	Slope	LCI	UCI	p value	Slope	LCI	UCI	p value	Slope	LCI	UCI	p value	Slope	LCI	UCI	p value
NBB	0.0018	0.0010	0.0026	< 0.001	0.018	-0.018	0.057	0.301	-0.019	-0.043	0.012	0.214	-4.71	-8.69	-1.65	< 0.001
CBB	0.0021	0.0014	0.0026	< 0.001	0.016	-0.004	0.039	0.079	0.009	-0.002	0.025	0.146	-3.42	-6.20	-1.43	< 0.001
SBB	0.0018	0.0014	0.0023	< 0.001	0.020	0.001	0.049	0.048	0.003	-0.007	0.017	0.660	-5.77	-10.77	-3.04	< 0.001

(TMDLs) calculated (Fla. Admin. Code, 2016). There is a lower threshold for surface waters that are placed on the state's Watch List and assessed more frequently, but do not require

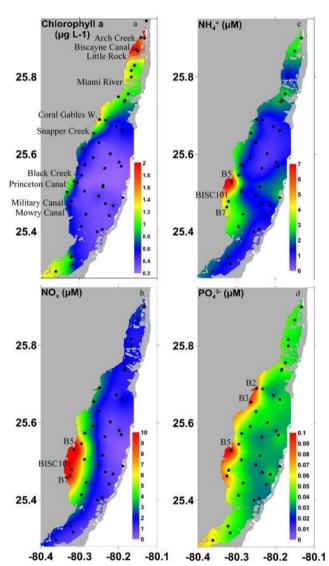


Fig. 5 The average concentration of **a** chlorophyll a (µg L⁻¹), **b** NO_x (µM), **c** NH₄⁺ (µM), and **d** PO₄³⁻ (µM) concentrations at individual stations throughout Biscayne Bay. Specific stations mentioned in the results and discussion are labeled on the maps

the development of TMDLs, which can be quite time and resource intensive. During the most recent assessment cycle, completed in 2017, all eight estuary nutrient regions (ENR) in Biscayne Bay were added to the Verified List for a TMDL. The Biscayne Bay ENRs were added because at least 2 of the 3 most recent annual geometric means of chlorophyll *a* were above the criteria set in each of the ENRs (Florida Department of Environmental Protection 2018).

While the current methodology used by the state of Florida identified Biscayne Bay as impaired, our approach of analyzing long-term trends at individual stations would improve the identification of impaired bodies of water and the efficiency of subsequent assessments and remedial actions. First, segmentation that averages multiple stations together results in the signal in the average being attenuated compared to the signal for stations with the strongest signal. This is especially pronounced in estuaries with a large number of runoff points, such as Biscayne Bay. The current minimum criteria for surface waters are determined within each ENR in Biscayne Bay. The criteria for Biscayne Bay are based on the conditions in a system that was already experiencing increases in chlorophyll a and PO_4^{3-} in most areas based on our analyses (Table 3, Fig. 6). Thus, it is impossible to apply this criterion to each individual station within the ENR. The heterogeneity within the ENR would suggest some stations would always be in exceedance and others would likely never exceed the criteria within the same ENR (Fig. 5).

Using the trends at individual stations rather than exceedance of a criteria allows for the identification of specific areas within an estuary that are degrading most quickly. This allows for more effective and efficient future action, especially in large estuaries with multiple watersheds and multiple runoff points. For example, all of Biscayne Bay is currently on the Verified List and required to have TMDLs developed for all runoff points into the Bay. Our individual station approach identifies areas where chlorophyll *a* and NO_x concentrations are increasing at a higher rate compared to the surrounding area. Since some of these areas are associated with specific canal outflows, this can aid in the development of an effective and targeted TMDL. Additionally, all nutrient impairments in Florida, including Biscayne Bay, are listed as a "medium"



Table 4 The annual rate of change of chlorophyll a concentrations ($\mu g L^{-1} \text{ year}^{-1}$) at individual stations throughout Biscayne Bay, the lower and upper 95% confidence intervals (LCI and UCI, respectively), and the p value. The annual rates of change were estimated using the Theil-Sen slope from the seasonal Mann-Kendall test. Italicized slope values refer to a slope at an individual station that is significantly different from the regional rate of change based on a lack of overlap between the 95% confidence intervals between the slopes

BB02 NBB 0.035 - 0.004 0.076 0.076 BB05A NBB 0.018 0.000 0.035 0.055 BISC134 NBB - 0.012 - 0.030 0.008 0.289 BB09 NBB 0.037 0.018 0.058 < 0.001 BISC133 NBB 0.046 0.023 0.066 < 0.001 BB14 NBB 0.018 0.010 0.030 < 0.001 BISC132 NBB 0.026 0.002 0.051 0.032 BB17 NBB 0.017 0.007 0.028 < 0.001 BISC131 NBB 0.011 - 0.002 0.027 0.110 BB22 NBB 0.012 0.003 0.021 0.013 BB54 CBB 0.030 0.009 0.068 0.013 BISC129 CBB 0.010 0.000 0.021 0.046 B2 CBB 0.044 - 0.004 0.118 0.078
BISC134 NBB - 0.012 - 0.030 0.008 0.289 BB09 NBB 0.037 0.018 0.058 < 0.001
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BISC131 NBB 0.011 -0.002 0.027 0.110 BB22 NBB 0.012 0.003 0.021 0.013 BB54 CBB 0.030 0.009 0.070 0.009 B1 CBB 0.042 0.009 0.068 0.013 BISC129 CBB 0.010 0.000 0.021 0.046 B2 CBB 0.044 -0.004 0.118 0.078 BISC128 CBB -0.006 -0.015 0.003 0.151
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B1 CBB 0.042 0.009 0.068 0.013 BISC129 CBB 0.010 0.000 0.021 0.046 B2 CBB 0.044 -0.004 0.118 0.078 BISC128 CBB -0.006 -0.015 0.003 0.151
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B2 CBB 0.044 -0.004 0.118 0.078 BISC128 CBB -0.006 -0.015 0.003 0.151
BISC128 CBB - 0.006 - 0.015 0.003 0.151
RISC126 CRR 0.001 = 0.009 0.011 0.974
DISC120 CDD 0.001 0.000 0.011 0.8/4
B16 CBB - 0.005 - 0.015 0.007 0.587
B3 CBB 0.039 0.016 0.086 0.003
BISC127 CBB - 0.002 - 0.006 0.003 0.595
BISC104 CBB 0.001 -0.002 0.004 0.652
B4 CBB 0.011 0.002 0.023 0.013
B15 CBB 0.004 -0.003 0.013 0.152
BISC103 CBB 0.014 0.008 0.019 < 0.001
BISC108 CBB 0.003 0.000 0.004 0.024
BB52 CBB 0.013 0.009 0.019 < 0.001
BISC109 CBB 0.005 0.001 0.008 0.014
BISC102 CBB 0.009 0.006 0.013 < 0.001
B5 SBB 0.014 -0.017 0.066 0.205
BB39A SBB 0.008 0.003 0.012 < 0.001
BB38 SBB 0.001 0.000 0.003 < 0.001
BISC111 SBB 0.004 0.002 0.006 < 0.001
BISC110 SBB 0.003 0.001 0.005 0.006
BB53 SBB 0.030 0.014 0.048 < 0.001
B6 SBB 0.020 0.005 0.026 0.007
BISC112 SBB 0.003 -0.0001 0.007 0.063
BISC101 SBB 0.008 0.005 0.011 < 0.001
BISC124 SBB 0.004 0.000 0.009 0.053
B14 SBB 0.013 0.004 0.024 0.002
B7 SBB 0.003 -0.011 0.018 0.884
BISC123 SBB 0.008 0.006 0.011 < 0.001
BISC113 SBB 0.006 0.002 0.010 0.008
BISC122 SBB 0.007 0.005 0.010 < 0.001
B8 SBB 0.015 0.009 0.022 <0.001

Table 4 (continued)

Station name	Region	Slope	LCI	UCI	p value
BISC116	SBB	0.006	0.001	0.010	< 0.001
BB47	SBB	0.011	0.008	0.018	< 0.001
B9	SBB	0.025	0.002	0.056	0.032
BISC135	SBB	0.020	0.011	0.029	< 0.001
B10	SBB	0.037	0.016	0.069	0.001

priory for TMDL development, meaning that a TMDL will be developed in the next 5 to 10 years (Florida Department of Environmental Protection 2018). However, the high rate of change of chlorophyll *a* concentrations in NBB (Fig. 6) suggests a need for more refined prioritization that ensures the development of TMDLs in certain ENR over before others.

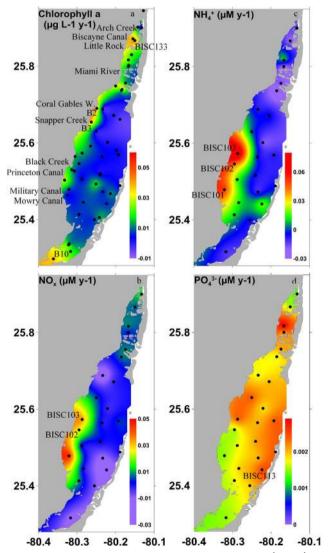


Fig. 6 The annual rate of change in **a** chlorophyll a (μ g L⁻¹ year⁻¹), **b** NO_x (μ M year⁻¹), **c** NH₄⁺ (μ M year⁻¹), and **d** PO₄³⁻ (μ M year⁻¹) concentrations at individual stations throughout Biscayne Bay. Specific stations mentioned in the results are labeled on the maps



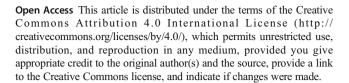
Our analysis showed that chlorophyll a and phosphate concentrations have been increasing at a high rate since 1996 throughout all of NBB. Moreover, there has been a recent dieoff of seagrass and subsequently slow recovery in NBB (Avila et al. 2017). Considering the seagrass die-offs and increases in chlorophyll a have been connected in other systems (Kemp et al. 1983; Twilley et al. 1985; Glibert et al. 2014), it is likely that the high and increasing chlorophyll a concentrations in NBB are somehow tied to the seagrass die-off (Zhang et al. 2003). The high rate of increase in chlorophyll a and seagrass die-off in NBB suggests the rapid decline in water quality is having an impact on the ecosystem, further demonstrating this region should have a higher priority listing compared to the rest of Biscayne Bay.

Overall, our analysis suggests that the State's identification of impaired water bodies can be improved to more appropriately differentiate levels of impairment. Even grouping the stations into sub-regions (ENRs) still masked the most impaired areas in CBB and SBB, which were smaller spatially than the sub-region scale. This has allowed some areas of Biscayne Bay to become substantially impaired before being added to the planning list for a TMDL. It is also reasonable to expect that water quality will continue to decline while a TDML is being developed. For NBB, 5–10 years is likely too long to wait for action to be taken.

The analysis of long-term trends at individual stations exposed a large degree of spatial variability not observed in the regional analysis (Tables 3 and 4). Through our analysis of individual stations, we were able to identify small-scale variability in the rate of change for chlorophyll a and nutrient concentrations, which allowed us to determine where water quality is deteriorating at the highest rate in Biscayne Bay (Fig. 6). The location of the highest rates of increase in chlorophyll a concentrations likely indicates that local sources of nutrients from the watershed are driving these long-term increases in chlorophyll a concentrations and degrading water quality in Biscayne Bay. The spatially explicit trends we discovered through studying the small-scale variability in longterm chlorophyll a and nutrient concentrations can be used by the state of Florida and local governments to improve the identification of impaired water bodies and implement highly targeted management strategies.

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