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Baseline

Widespread sewage pollution of the Indian River Lagoon system, Florida (USA) resolved by spatial analyses of macroalgal biogeochemistry

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ABSTRACT

The Indian River Lagoon (IRL) system, a poorly flushed 240 km long estuary in east-central Florida (USA), previously received 200 MLD of point source municipal wastewater that was largely mitigated by the mid-1990's. Since then, non-point source loads, including septic tank effluent, have become more important. Seventy sites were sampled for bloom-forming macroalgae and analyzed for $\delta^{15}N$, % nitrogen, % phosphorus, carbon:nitrogen, carbon:phosphorus, and nitrogen:phosphorus ratios. Data were fitted to geospatial models showing elevated $\delta^{15}N$ values (> +5‰), matching human wastewater in most of the IRL system, with elevated enrichment ($\delta^{15}N \geq +7$‰ to +10‰) in urbanized portions of the central IRL and Banana River Lagoon. Results suggest increased mobilization of OSDS NH_4^+ during the wetter 2014 season. Resource managers must improve municipal wastewater treatment infrastructure and commence significant septic-to-sewer conversion to mitigate nitrogen over-enrichment, water quality decline and habitat loss as mandated in the Tampa and Sarasota Bays and the Florida Keys.

Coastal estuaries, world-wide and in particular, the United States, have undergone significant ecosystem decline as a result of human population growth and cascading ecological impacts (Nixon, 1995; NRC, 2000; Bricker et al., 2007). This follows recognition that urbanization and agricultural alteration of formerly natural watersheds has resulted in unsustainable nutrient over-enrichment with water quality decline, harmful algal blooms, habitat loss, and loss of fisheries being well reported symptoms of ecosystem decline and collapse (Nixon, 1995, Vitousek et al., 1997, NRC, 2000).

For estuaries of the southeastern United States, researchers predicted that estuarine ecosystems are not sustainable under the strain of human-induced land-use alterations of watersheds and subsequent water quality and habitat decline (Dame et al., 2000). Specifically, the NOAA National Estuarine Eutrophication Survey in 1997 (NOAA, 1997) indicated that the Indian River Lagoon (IRL) system, along east-central Florida (see Fig. 1), was "hypereutrophic" with respect to excessive carbon fixation (i.e. elevated chlorophyll-a, macroalgal blooms) resulting from nutrient over-enrichment. Indications of excess macroalgal biomass, and precipitous loss of seagrass coverage adjacent to urbanized portions of the watershed have been reported since the mid-1980's (see Virnstein and Carbonara, 1985, Virnstein, 1999, Barile and Lapointe, 1999). In 2011, the northern IRL's "superbloom" was a predictable ecological phase-shift (see Valiela et al., 1992) to a toxic phytoplankton-dominated system (Phlips et al., 2004), which followed the loss of seagrass and benthic macroalgae (SJRWMD, 2012) and occurred, simultaneously, with die-offs of manatee, dolphins, seabirds, and periodic fish kills. However, before the "superbloom" event, toxic phytoplankton blooms had already become a common feature of the IRL system. For example, the toxin producing dinoflagellate species *Pyrodinium bahamense* var. *bahamense* (a saxitoxin producer, see Landsberg, 2002, Landsberg et al., 2006), and other HAB species, such as the diatom *Pseudo-nitzschia pseudodelicatissima* have been reported in bloom concentrations (Phlips et al., 2002, Phlips et al., 2004, Phlips et al., 2006). More recent brown tide outbreaks in the central and northern portion of the system have degraded water quality causing widespread water column oxygen deficits and subsequent fish kills (Gobler et al., 2013, Kang et al., 2015). These ecological indicators have prompted recognition of the role of escalating nutrient over-enrichment as a driver of ecosystem collapse (see Barile and Lapointe, 1999; Lapointe et al., 2015); and mandates by state and federal regulatory agencies for mitigation for land-based nutrient sources loadings into the IRL system.

Not unlike many estuaries around the United States, the watershed of the Indian River Lagoon system has undergone significant land-use alteration (IRL NEP, 1996). Over the past 100 years, the watershed transitioned from a natural wetland with marginal human inhabitation to an agrarian and now growing urban land-use. Because of the long expanse (~240 km), the Indian River Lagoon system has been biogeographically divided into 5 sub-sections to include: the Mosquito Lagoon to the north, the Banana River lagoon to the east, and the northern,

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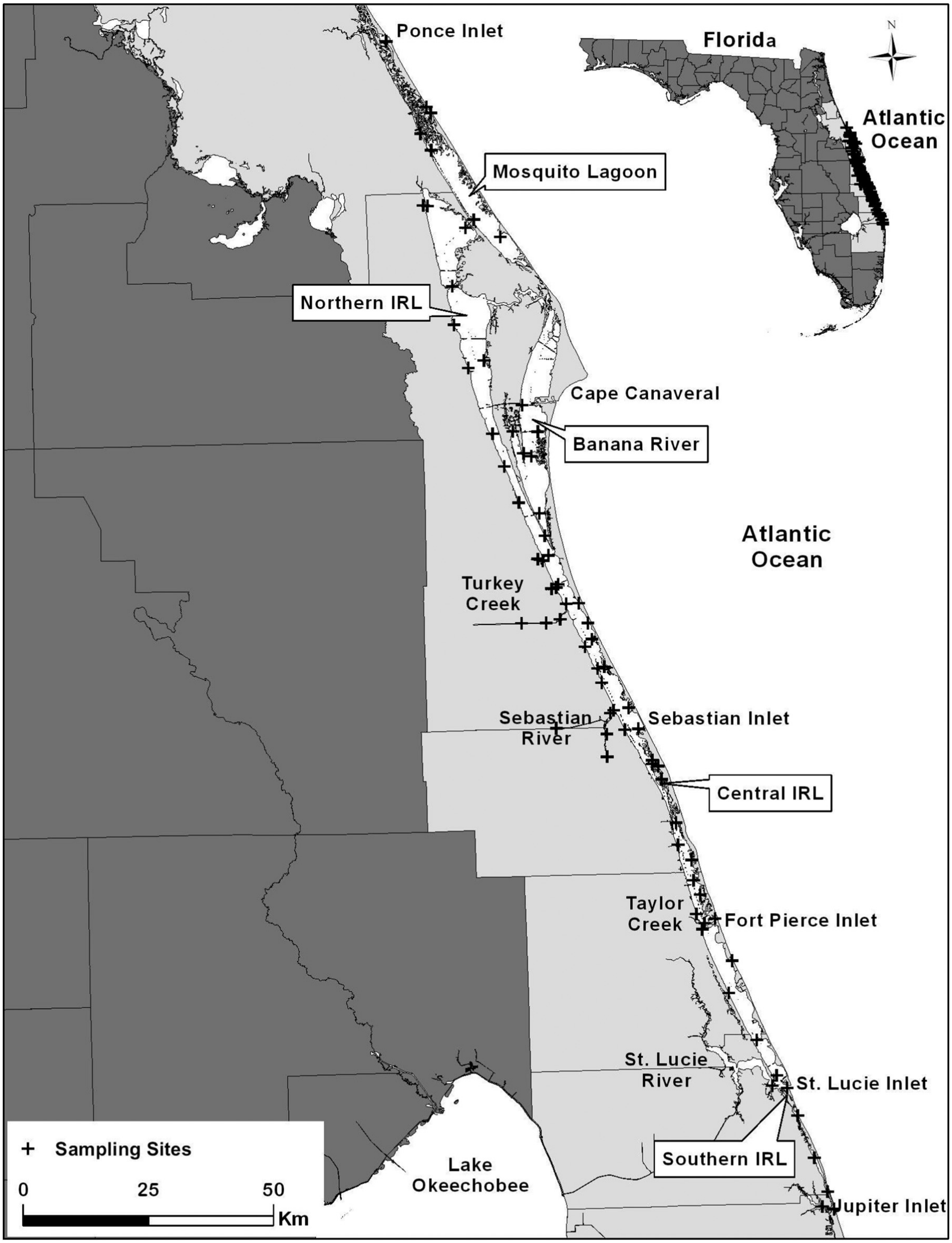


Fig. 1. Map of the Indian River lagoon system. Included are labels for the 5 subsections of the IRL system: Mosquito Lagoon, Banana River Lagoon, Northern, Central and Southern Indian River Lagoon. Sample site locations denoted as (+).

central, and southern portions of the IRL (see Fig. 1).

Panama canal-era (1920‘s) construction of 100’s of kilometers of upland drainage canals converted nearly 1800 km^2 of wetlands to farmlands that provided a surface and groundwater conveyance system downstream in the central and southern portions of the Indian River Lagoon system. This development of upland drainage canals, which drained wetlands formerly flowing west to the St. Johns River and Lake Okeechobee basins, expanded the watershed by three times to nearly 600, 000 ha, particularly in the southern portions of the IRL system watersheds. Alternatively, the central IRL watershed is the most urbanized with ~450 people per square kilometer (Sigua et al., 2000), versus the more sparsely populated northern IRL and Mosquito Lagoon sub sections.

Table 1

Summary data of wastewater treatment method for residences in counties along the Indian River Lagoon estuary system comparing onsite sewage disposal system (OSDS, septic and package plant systems) and municipal wastewater treatment (Municipal) usage. Data from Florida Department of Health's Florida Water Management Inventory Project. http://www.floridahealth.gov/environmental-health/onsite-sewage/research/flwmi/index.html

	Wastewater treatment method		
County	OSDS	Municipal	Proportion OSDS
Volusia	102,831	102,413	50%
Brevard	91, 630	117,797	43%
Indian River	30, 574	25,968	54%
St. Lucie	34,364	70,649	33%
Martin	29,864	26,201	53%
N. Palm Beach (City of Jupiter)	3326	34,048	10%

1. Sources of nutrient enrichment to the Indian River Lagoon system

The narrow and poorly-flushed IRL system (Smith, 1993) has experienced nitrogen and phosphorus over-enrichment at levels exceeding concentration thresholds for restoration of sub-tropical seagrass ecosystems (see Sigua et al., 2000; Tomasko and Lapointe, 2001), a keystone habitat component. The Florida Department of Environmental Protection (FDEP, 2013) adopted Total Maximum Daily Loads (TMDL) for 3 basins (sub-sections) of the IRL system, with respective Basin Management Action Plans (BMAP) designed to reduce loads of nitrogen and phosphorus loading to the IRL system. Described below, are significant "external" macronutrient input sources from the airshed, and surface and groundwaters from the watershed and their potential loading contributions to the IRL system, from the technical literature supporting the IRL BMAP.

2. Atmospheric deposition

Estuaries downgradient from urban areas, particularly along eastern continental fringes, receive significant airshed loadings of reactive nitrogen (Barile and Lapointe, 2005). The Indian River Lagoon system is no different. National Atmospheric Deposition Program site (NADP, FL-99, wet + dry deposition) between the northern IRL and Banana River at Kennedy Space Center, and an EPA supported wet deposition site in the central IRL region at Sebastian Inlet, were used to model a direct IRL reactive nitrogen deposition rate of 3.7 kg/ha. For the northern portion of the IRL where riverine input is at a minimum, this deposition rate may represent ~25% of the reactive nitrogen loading. In the central and southern portions of the IRL system, the contribution of this source is significantly less at < 10% (Janicki Environmental and Applied Technology and Management, 2012).

3. Agricultural and urban fertilizer

Agricultural and urban fertilizer broadly represents the most significant nitrogen and phosphorus input sources to adjacent surface water systems in Florida (Badruzzaman et al., 2012). In east central Florida, agricultural (citrus, row crops and cattle) operations represent ~50% of the southern IRL system watershed, which is hydrologically-linked by extensive drainage systems down gradient to the IRL system (Graves et al., 2004). Nitrogen losses have been reported to be significant in agricultural areas within the southern IRL watershed. Increased mobilization of nitrogen is related to rain intensity, soil nitrogen level, and rate of fertilizer application, with citrus and vegetable farms contributing the highest nitrogen loads (Zhang et al., 2007). Agriculture operations in the watersheds of the Northern Everglades and southern IRL system, including the St. Lucie estuary, are under order (Florida Statutes Section 373.4595(2) for nutrient load reduction BMAPs. These BMAPs have been adopted by nearly 90% of agricultural operations in the southern IRL system watershed and have been effective at removal of ~600 metric tons of nitrogen from the St. Lucie & Lake Okeechobee watersheds that may discharge into the southern IRL (FDACS, 2016).

Extensive residential fertilizer use in urban areas with a population of > 1 million residents along the IRL system has become an important nitrogen and phosphorus input to residential lawns, as population has increased in urbanized watersheds of the system. Two significant regulatory actions have significantly reduced both the application rates and hence the loading of urban fertilizers to the IRL system. First, the Urban Turf Fertilizer Rule effective December 31, 2007 (5E-1.003 F.A.C.) banned the use of phosphorus from residential fertilizers, and restricted application rates of both nitrogen and phosphorus. Also, preceding this study in 2013, the majority of counties and municipalities along the IRL system had adopted the State of Florida (FDEP, 2009) "Model ordinance for fertilizer use in urban landscapes" (Florida statutes, section 403.067). This ordinance is designed to mitigate fertilizer misapplication and potential loading to adjacent ground and surface water systems, as the ordinance "promotes the proper use of fertilizers by applicators, specifies application rates and methods, and prohibits application less than 15 feet from waterbodies, and establishes a rainy season (June through September) ban on application." Data from Brevard County (FDACS, 2015) indicate an ~80% decrease in fertilizer purchases preceding this study period (~1600 down to ~300 tons nitrogen/yr.), with the insinuation that decreases in purchase rates resulted in decreases in application rates in urban residences along the IRL watershed.

4. Onsite sewage disposal systems (OSDS)

The Florida Department of Health has inventoried residences utilizing OSDS versus municipal wastewater treatment for the 6 counties bordering the IRL system (see Table 1, Fig. 2). In most of the counties reliance on OSDS is high, often near 50% of the residences. Lapointe and Krupa (1995a, 1995b), Lapointe et al. (2012, 2015, 2017) has demonstrated significant contamination of high density OSDS effluents through adjacent ground and surface waters into tributaries of the IRL system. OSDS systems are often in high densities in residential communities in the watershed, sited upon geological units with high hydraulic conductivities either upgradient along the western border of the Lagoon, or on siliciclastic (sand) barrier islands (see Fig. 2). Groundwater and subsurface discharge may contribute 45 to 60% of the total nitrogen and total phosphorus loading to the IRL system (see Janicki Environmental and Applied Technology and Management, 2012); the evidence suggests OSDS effluents are an important component of this loading.

5. Wastewater treatment plant discharges

Although municipal wastewater treatment plant systems (WWTPs) operate under strict performance guidelines under federal (USEPA) and state (FDEP) regulatory programs, they have historically been placed alongside the Lagoon system to allow for quick and efficient surface water discharge during excessive rainfall events. The Indian River Lagoon Act in 1990 banned direct surface water discharge of secondarily-treated human wastewater effluents that had escalated to ~200 million liters per day (Sigua et al., 2000). However, regulations continue to permit up to 90 days per year of "emergency wet weather" surface discharge into the IRL system, when significant rain events overload treatment system capabilities (Florida Department of Environmental Protection (FDEP), 2017). The impact of these discharges may be less significant in the southern portion of the IRL system where lower residence time of effluents, and hence flushing from the system, result from the proximity of WWTPs to oceanic inlets. With the

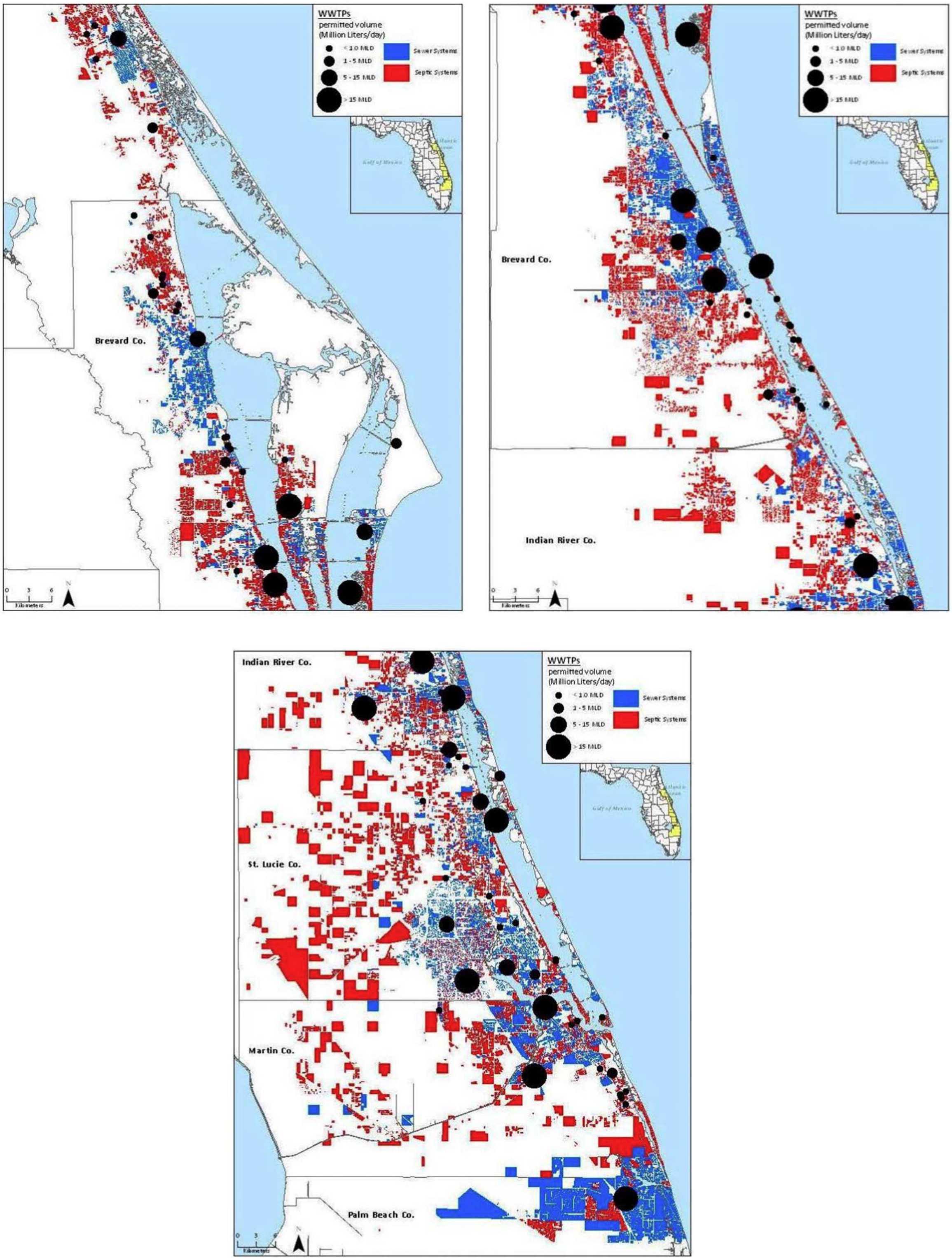


Fig. 2. Geospatial map of human wastewater treatment methods in the Indian River Lagoon region. Municipal wastewater treatment plants are denoted in black dots based upon their permitted treatment capacities, from FDEP (2017). Residence served by municipal wastewater treatment are denoted in blue, residences utilizing onsite sewage disposal systems are denoted in red. Data from Florida Department of Health's Florida Water Management Inventory Project. http://www.floridahealth.gov/environmental-health/onsite-sewage/research/flwmi/index.html (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

emerging significance of non-point nutrient loading to estuaries in recent decades, this point source has been reduced to an average of ~5% of the nitrogen and phosphorus loading in the northern IRL system, but may be on average higher (~10%) in the Banana River segment (Janicki Environmental and Applied Technology and Management, 2012). However, these estimates do not account for significant but poorly reported "wet weather" discharges (FDEP, 2017, Thacker, 2004); in some cases, several million liters per day per treatment plant during wet season events that may be important for supporting harmful algal blooms. Municipal wastewater treatment facilities are largely

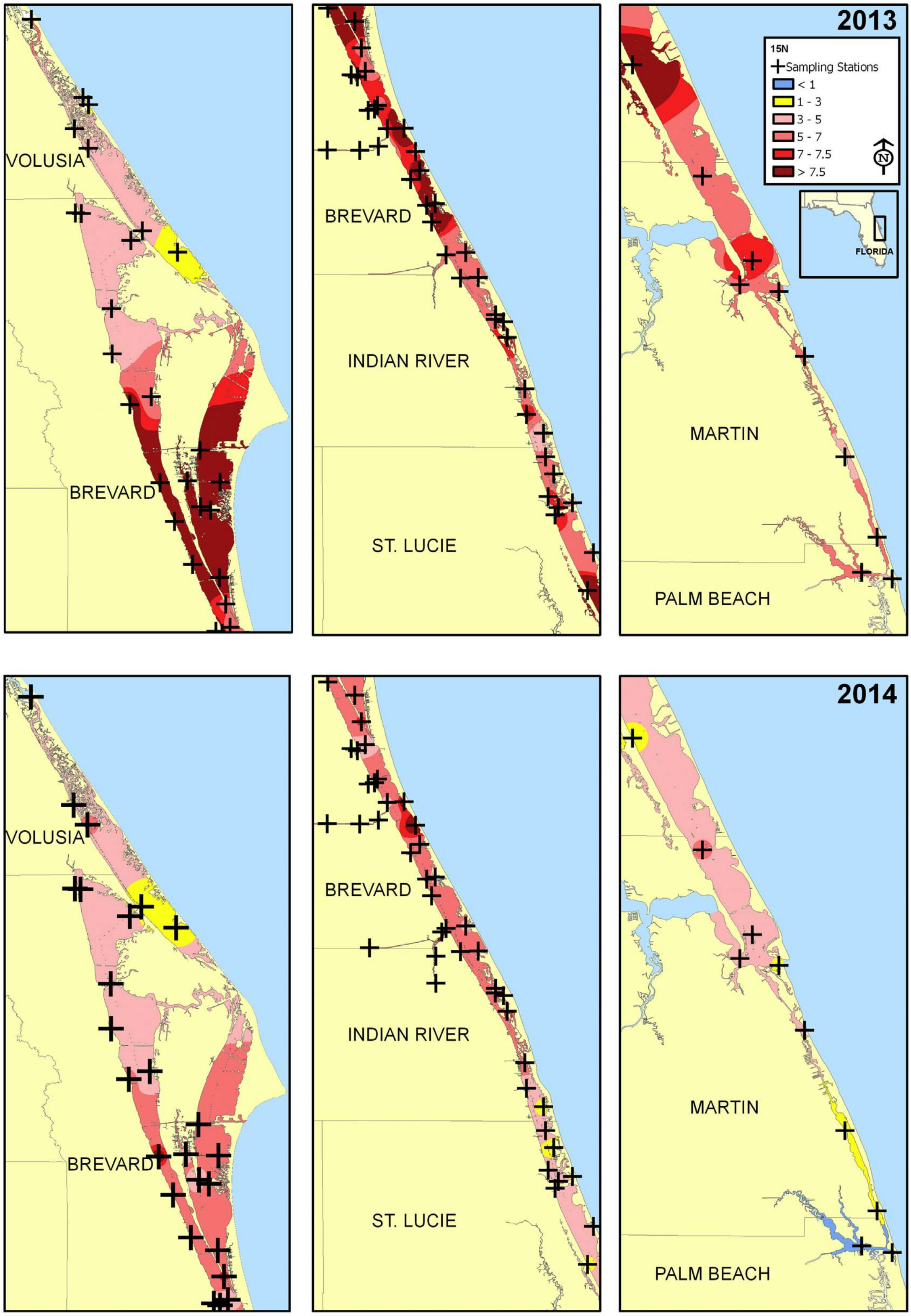


Fig. 3. Geospatial map of $\delta^{15}N$ (‰) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

situated along the IRL system to facilitate wet event discharges to surface waters (see Fig. 2).

The most northern IRL system sub-section, the Mosquito Lagoon (ML) in Volusia County, is heavily urbanized with municipal WWTPs to the north and on the barrier island; and receives regular tidal flushing from Ponce Inlet to the north. But the more sparsely populated portion in the central ML is largely dependent upon residences with OSDS along the Lagoon. The Banana River Lagoon (BRL) portion of the Indian River Lagoon system, is relatively enclosed and surrounded by several municipal wastewater treatment plants (WWTPs), one that has a direct surface water discharge permit of ~400,000 L/day (FDEP, 2017). Although less urbanized, the restricted Kennedy Space Center/ Canaveral Air Force Station to the north is serviced by centralized WWTPs. The middle and lower portions of the northern IRL have high volume

WWTPs serving municipal areas, but heavy reliance upon OSDS, particularly adjacent to the Lagoon in the southern portion of the subsection, on both the mainland and the thin barrier island between the N-IRL and the BRL. The central and southern portions of the BRL have urbanized residential land-use that relies largely upon OSDS on the western and southern upland area. Whereas the eastern barrier island is largely serviced by municipal WWTPs. The heavily urbanized central IRL sub-section has extensive municipal wastewater treatment, except for the City of Palm Bay, which has nearly 30,000 residences utilizing OSDS, located beyond the service boundary for municipal wastewater treatment. Additionally, there is significant reliance on OSDS on portions of the barrier island between the BRL and central IRL, and the southern Brevard County barrier island. The southern IRL sub-section has municipal WWTPs in some municipalities, but significant reliance on OSDS in urban areas of southern St. Lucie County as well as the majority of the urbanized and barrier island portions of Martin County (see Fig. 2).

6. Biogeochemical tracers of nutrient sources

Bio-indictors provide a reliable method to assess the temporal and spatial dynamics of nutrient enrichment processes in coastal water bodies. Sessile marine macroalgae, specifically, function as continuous sampling monitors for nutrient availability with their tissue biochemistry serving as an integrative record of water column nutrient chemistry over extended periods of time (Levine, 1984). As a result, macroalgal tissue contents are routinely used to discriminate sources of nitrogen enrichment to coastal waterbodies using natural abundances of stable nitrogen isotopes (see review by Risk et al., 2009), and the many examples using this method to track anthropogenic nitrogen sources in coastal macroalgae (McClelland and Valiela, 1998, Costanzo et al., 2001, Barile, 2004, Savage and Elmgren, 2004, Lapointe et al., 2005, Barile and Lapointe, 2005, Cohen and Fong, 2005, Dailer et al., 2010). This method has also proven useful for discriminating multiple anthropogenic nitrogen signals between atmospheric deposition, fertilizers, and human wastewater (see McClelland et al., 1997). Further, human wastewater from municipal treatment systems can be discriminated from OSDS (Steffy and Kilham, 2004). Other wastewater sources, such as Class V sewage injection wells (Dailer et al., 2010) and ocean outfalls (Lapointe et al., 2005) and multiple nitrogen species associated with OSDS (Lapointe et al., 2017) have distinct nitrogen isotope signatures.

In addition, macroalgal tissue nutrient concentrations of carbon, nitrogen and phosphorus and their ratios (C:N:P) can provide temporal and spatial information primary nutrient availability from respective land-based sources, as macroalgae are known to store excess available inorganic nitrogen and phosphorus from the water column.

Biochemical data from algae tissue bioindicators were compared along spatial gradients with respect to adjacent land-uses and primary nutrient sources that varied between the 5 sub-sections of the IRL system. Overall, general spatial trends in nitrogen and phosphorus availability between these sub-sections have been reported elsewhere (Sigua et al., 2000; Lapointe et al., 2015). Reported here, are fine-scale comparisons with adjacent land-uses, and comparisons between the 2013 & 2014 wet seasons. The ArcGIS Spatial Analyst package produced spatial models of interpolated δ15N values (Fig. 3), changes in mean δ15N values between 2013 and 2014 (Fig. 4), %N (Fig. 5), %P (Fig. 6), C:N (Fig. 7), C:P (Fig. 8) and N:P (Fig. 9) to provide spatial discrimination along the IRL system, and a comparison between the distinct 2013 and 2014 wet seasons. Lastly, δ15N values for macrophytes for the 2014 wet season in two upland tributaries of the central IRL, Turkey Creek and Sebastian River, are plotted versus adjacent OSDS and municipal wastewater treatment uses in Fig. 10.

7. Study area and sample collection

A spatially explicit grid of seventy sample sites was established across the 5 sub-sections of the Indian River Lagoon system extending from the Mosquito Lagoon, near Ponce Inlet (New Smyrna Beach) ~240 km to the southern IRL at Jupiter Inlet (see Fig. 1). At each sample site, at least 2 attached macroalgae species were collected for biochemical analyses during the wet season of September to October in both 2013 and 2014. Included in this assessment are sample sites in upland tributaries of the Indian River Lagoon that are potential nutrient sources to the IRL. However, the southern Lagoon tributary, the St. Lucie River estuary which receives discharges from Lake Okeechobee, was excluded from this analysis as it has been subjected to intensive assessment in previous reports (see Lapointe et al., 2012, Lapointe et al., 2017). At sample sites in upland freshwater tributaries, bloom forming freshwater algae and aquatic plants were collected and utilized for analyses.

To characterize the hydrologic conditions of the two sample years, 2013 and 2014, NOAA National Weather Service precipitation data from 6 locations along the Lagoon were compiled. Because the magnitude of rainfall, over time, is a driver of both stormwater and groundwater baseflow to the Lagoon system, flux of both nitrogen and phosphorus results during varying hydrological conditions. For example, historically significant rain events during the September to October period, associated with monsoonal rain patterns and tropical system events, result in significant “emergency” WWTP sewage discharges and significant urban and agricultural freshwater discharges to the estuary by water management authorities (i.e. South Florida and St. Johns Water Management Districts). As such, comparisons of nitrogen and phosphorus in macroalgal bioindicators were made between the 2013 and 2014 rainy seasons. In Table 2, below, rainfall totals from 6 meterological stations along the IRL system for 2013 and 2014 are reported below as 1) annual totals, and 2) antecedent “wet season” (July through September) totals preceding sampling.

8. Biogeochemical analyses

At each sample station, attached macroalgal species were collected for biochemical analyses. For δ^{15}N analysis, conspicuous attached macroalgal bloom species from the IRL region, preferentially those common to most sample sites allowed comparisons over spatial gradients and between years. The most common species collected, as a function of percentage from the total sample sites were: *Gracilaria tikvahiae* (84%), *Acanthophora spicifera* (42%), *Chaetomorpha* sp. (41%), *Ulva intestinalis* (15%), *Lyngbya majuscula* (14%) and *Caulerpa sertularioides* (13%). Cohen and Fong (2005) reported that macroalgal bloom species, such as *Ulva intestinalis*, do not show significant or inconsistent “fractionation effects” in estuarine waters as both water column dissolved inorganic nitrogen concentrations and macroalgal δ^{15}N values increase, and δ^{15}N values do not vary significantly among macroalgal functional forms. As such, they are conservative and reliable tracers of known δ^{15}N values from adjacent anthropogenic and natural nitrogen sources.

Composite samples of each macroalgae species were dried in a laboratory oven (60 °C) to constant weight, and dried samples were pulverized to a powder by mortar and pestle. Samples were then analyzed at the University of Georgia- Stable Isotope Ecology lab in a Carlo-Erba N/A 1500 elemental analyzer using Dumas combustion and the purified nitrogen gas was then measured by a VG Isomass mass spectrometer. The standard used for stable nitrogen isotope analysis was N_2 in air. δ^{15}N values, in per mil (‰) concentration were calculated as $[(R_{sample}/R_{standard}) - 1] \times 10^3$, with R equal to $^{15}N/^{14}N$. Analysis for total phosphorus content from respective samples was achieved by dry ash acid extraction following combustion of tissue samples in a muffle furnace at 500 °C, followed by atomic adsorption spectrophotometry (UGA-SIL, 2015a, 2015b), and percent weight was converted to molar

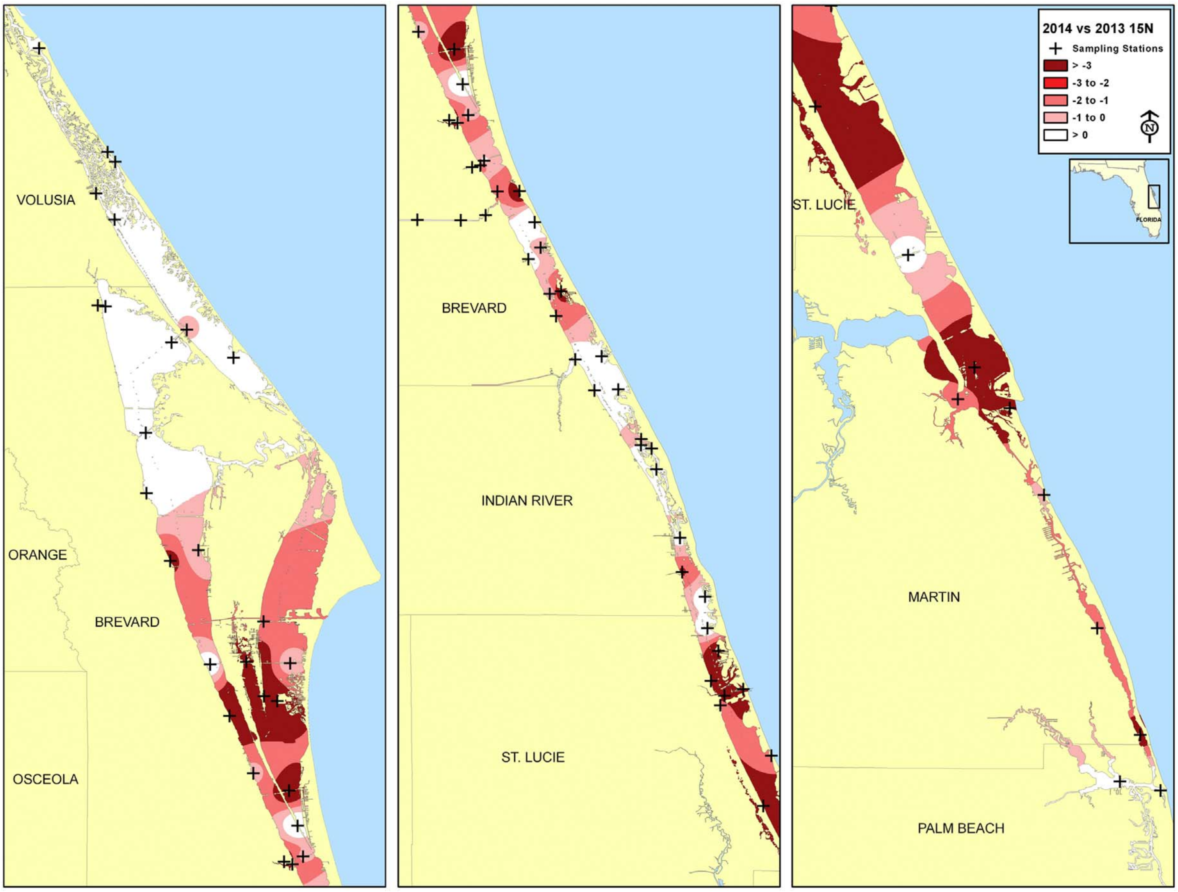


Fig. 4. Geospatial map of changes in $\delta^{15}N$ (‰) values in macroalgae in the Indian River Lagoon system between 2013 and 2014.

weight. Percent carbon, nitrogen weight contents from the element analysis preceding nitrogen and carbon isotope analysis were converted to molar equivalents of nitrogen and carbon, and were used to calculate molar ratios of carbon to nitrogen, carbon to phosphorus and nitrogen to phosphorus. Ratios of C:N:P from macroalgal tissue were presented spatially with respect to a modified Redfield ratio of 360:30:1 for macroalgae to characterize temporal and spatial variation in tissue nutrient status, where $C:N > 12$ represents N-limitation, $C:P > 360$ represents P-limitation, $N:P > 30$ represents P- limitation, and $N:P < 30$ represents N-limitation (Atkinson and Smith, 1983).

9. Spatial and statistical analyses

Average tissue values of $\delta^{15}N$, %N, %P, C:N, C:P and N:P for each site were fitted to a spatial model using the Spatial Analyst application of ArcGIS (2016) across the Indian River Lagoon system. Coefficients of variance for spatial models were < 25%, to assure coherence to the source data. Following *a priori* tests for homogeneity of variances, pairwise comparisons were made using non-parametric statistical comparisons. Annual (2013 versus 2014) values were compared with *t*-tests for the respective 5 sub-sections of the IRL system. Wastewater treatment plants (WWTPs) in the IRL watershed were plotted as a function of treatment capacity (see Fig. 2) to denote areas where municipal wastewater treatment versus onsite sewage disposal systems (OSDS, i.e. septic tanks and package plants) relative to macroalgal tissue collection sample sites. For municipal and private WWTPs along the Indian River Lagoon system, Florida Department of Environmental Protection (FDEP, 2016) database records for permitted treatment facilities were utilized for treatment type, volume of wastewater treatment and spatial location. FDEP (2017) permit records for these facilities were consulted to resolve daily permitted surface water discharge rates, emergency spills, discharges, etc. For OSDS, the Florida Department of Health Florida (FDOH) Water Management Inventory GIS database was imported to spatially indicate residences with active OSDS systems.

10. Comparison of $\delta^{15}N$, %N, %P, C:N, C:P, N:P values between 2013 and 2014

Across the spatial scale of the study area, the antecedent conditions in the wet season (July through September) preceding sampling, included significantly more rainfall (see Table 2) at 5 of the 6 meteorological stations reported along the IRL system. With increasing 2014 wet season rainfall, mean $\delta^{15}N$ values from the 2014 wet season declined (Fig. 4) significantly in macroalgae tissue pooled from 4 of the 5 sub-sections of the IRL system (Table 3). Tissue nitrogen contents (%N) increased significantly in 2 of the 5 sub-sections (see Table 3) in the wetter 2014 season. For %P, C:N and C:P values, there was no significant difference ($p > 0.05$) for all sub-sections of the IRL system between years. For N:P contents, only the central IRL showed significant ($p < 0.001$) differences between 2013 and 2014, and this was largely as a result of higher %N contents in 2014 in the central IRL subsection.

11. Mosquito Lagoon

Macroalgal $\delta^{15}N$ values averaged +3 to +5‰ at sites along much of the northern and central ML (Fig. 3). These values are consistent between 2013 and 2014 for most of the ML, except at the Oak Hill south sample site, which is directly downgradient of a low-lying community relying exclusively on OSDS for wastewater treatment. In 2014, values at that site were slightly enriched from +3 to +5‰ to +5 to +7‰, or a potentially more nitrified OSDS nitrogen source. At the southern end

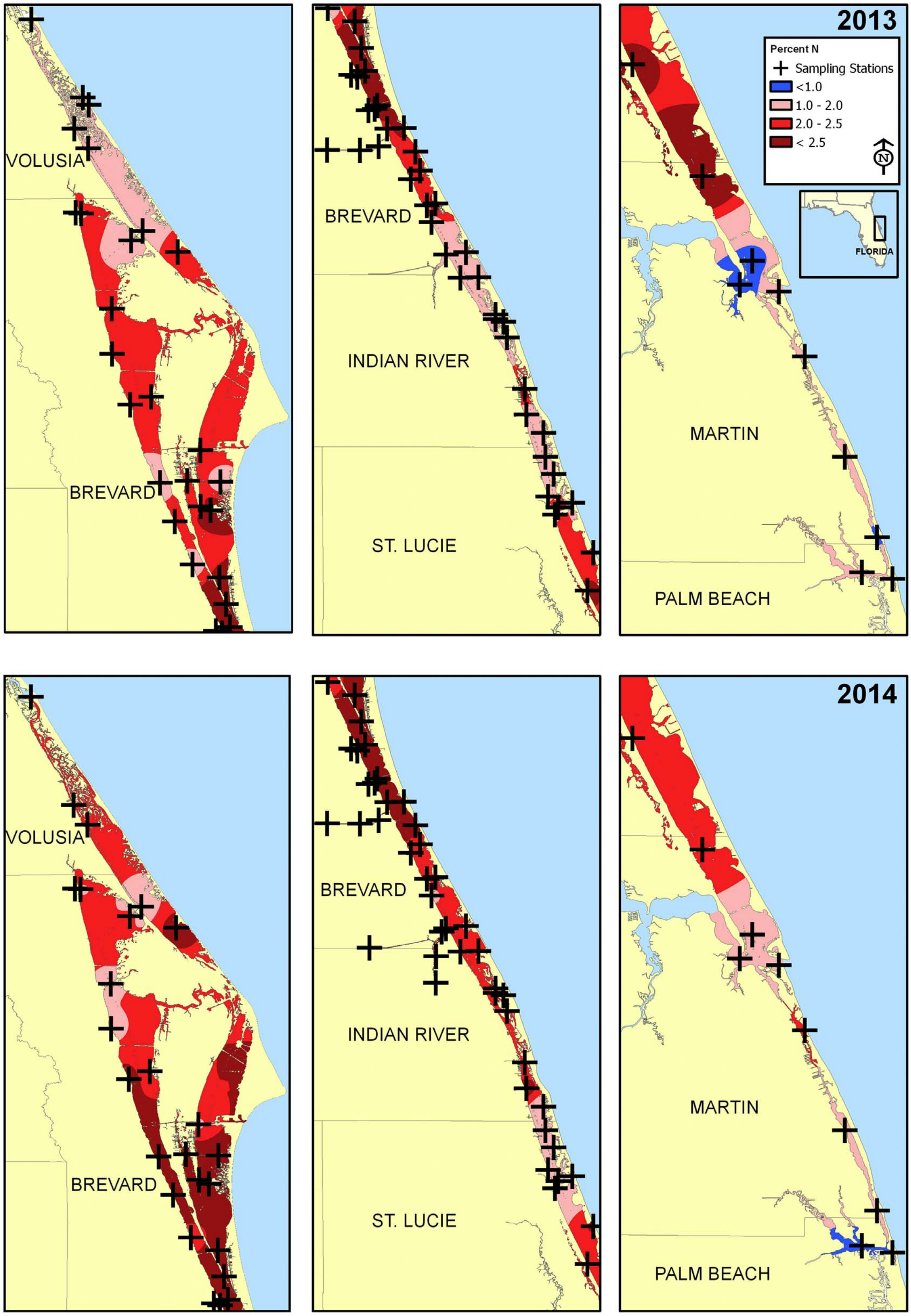


Fig. 5. Geospatial map of percent nitrogen (%N) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

of the Mosquito Lagoon, that is surrounded by the federally protected Canaveral National Seashore to the east and Merritt Island National Wildlife Refuge to the south, values of +1 to < +3‰ for both years are indicative of a background atmospheric nitrogen deposition or natural nitrogen fixation source.

Significantly higher tissue nitrogen contents are present in macroalgae during the wetter 2014 versus dryer 2013 (2.1% ± 0.4 versus 1.7% ± 0.3, $p < 0.05$). Phosphorus availability is generally very limited in this portion of the IRL system, with %P values largely below 0.2%, and not significantly different ($p > 0.05$) between years. High N:P ratios of > 40 were common between years throughout the study area except for a site with potential influence of natural bird rookeries at the east end of Haulover canal, a man-made inlet between Mosquito Lagoon and the Northern Indian River Lagoon (Fig. 9).

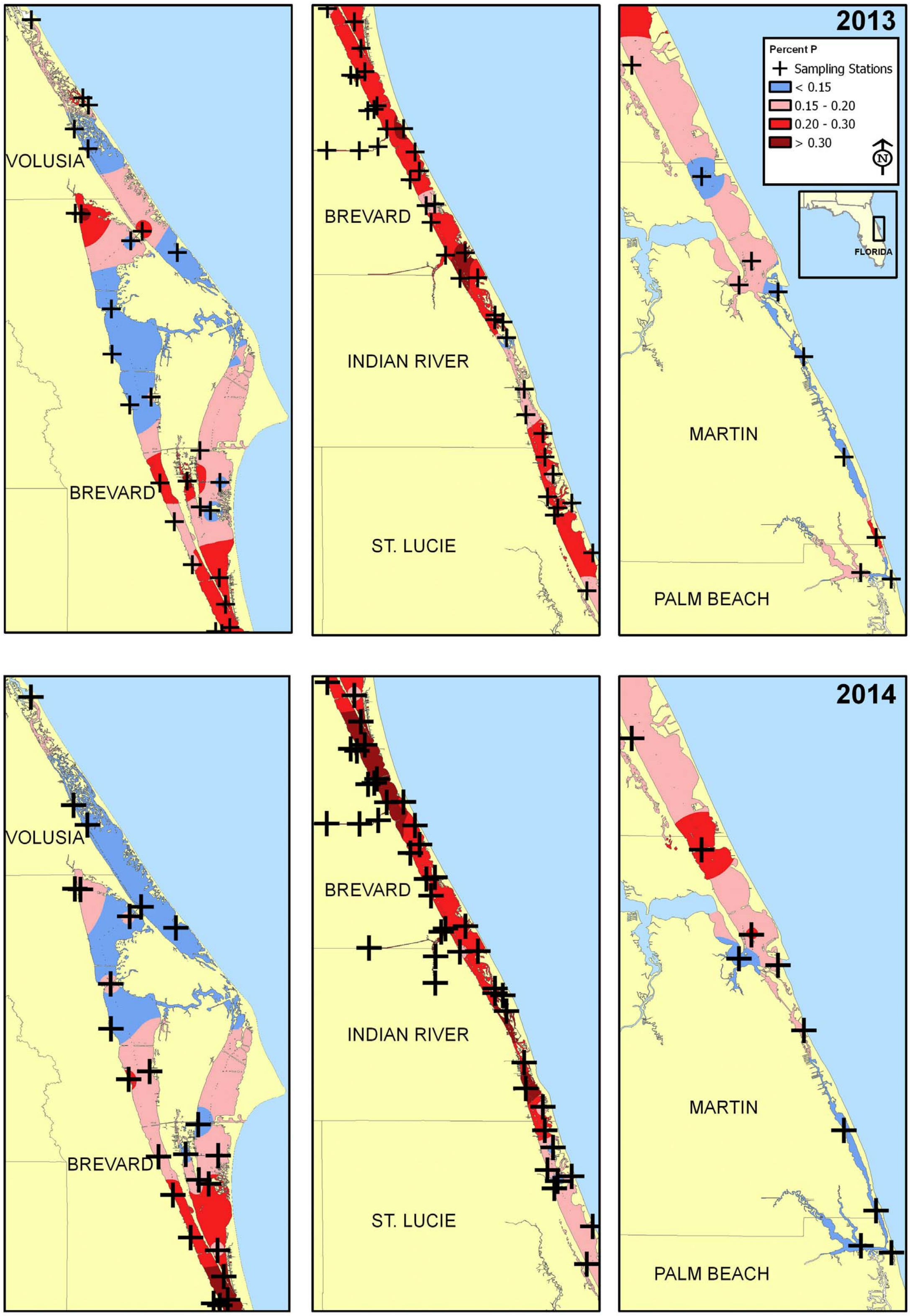


Fig. 6. Geospatial map of percent phosphorus (%P) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

12. Banana River Lagoon

In the drier 2013 sample year, $\delta^{15}N$ values were significantly higher in the Banana River Lagoon (BRL, see Table 2) and decreased by ~ +3‰ in the wetter 2014 year (Fig. 4). In 2014, values at 2 sites on the western portion of the BRL, lower $\delta^{15}N$ values of +3–5‰ were present adjacent to residential areas with high OSDS density. Whereas, values in the broader BRL system were higher at +5–7‰.

Percent tissue nitrogen values were high in both years (2.4% ± 0.6 versus 2.7% ± 0.4, but not significantly different), with highest values > 2.5% on average in the southern urbanized portion of the BRL. Variability in %P availability, likewise increased in a gradient, in both 2013 to 2014, from lower to higher concentrations (< 0.15% to > 0.3%) from north to south in the BRL. In the 2014 wet season,

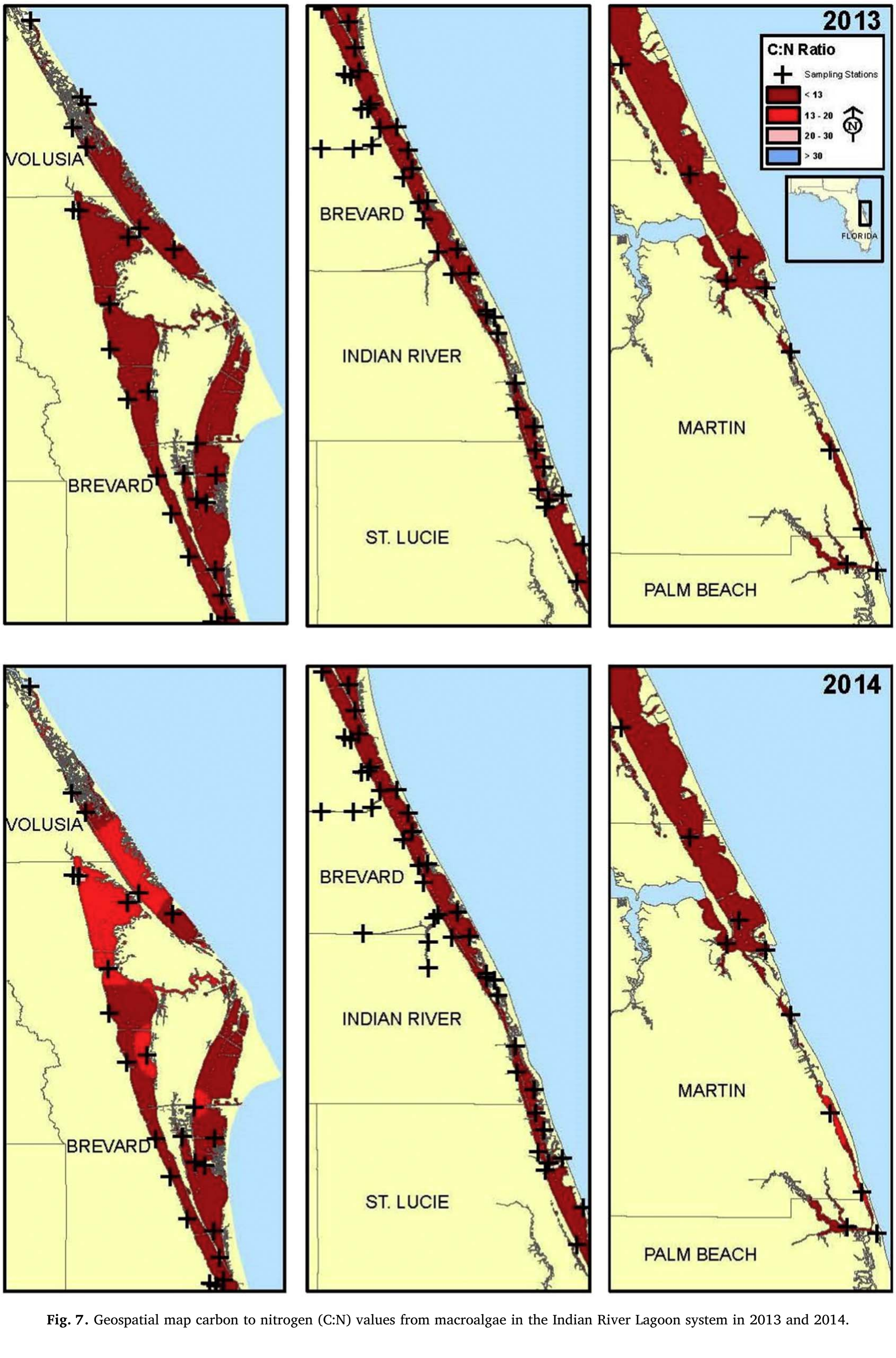


Fig. 7. Geospatial map carbon to nitrogen (C:N) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

values increased in the middle BRL adjacent to the Cocoa Beach WWTP. N:P ratio values of > 40 were present throughout the wetter 2014 season, with more balanced values of nitrogen versus phosphorus availability in the dryer 2013 season (N:P = 15 to 30) in the middle and lower portions of the BRL, although the overall mean N:P was not significantly different between 2013 and 2014 in BRL.

13. Northern Indian River Lagoon

In the northern IRL sub-section, significantly higher $\delta^{15}N$ values (> +7.5‰) were present in the dryer 2013 year in the heavily urbanized middle and lower portions of the sub-section; versus significantly lower (+5–7‰) values at sites in the wetter 2014 year (see Table 2, Fig. 4).

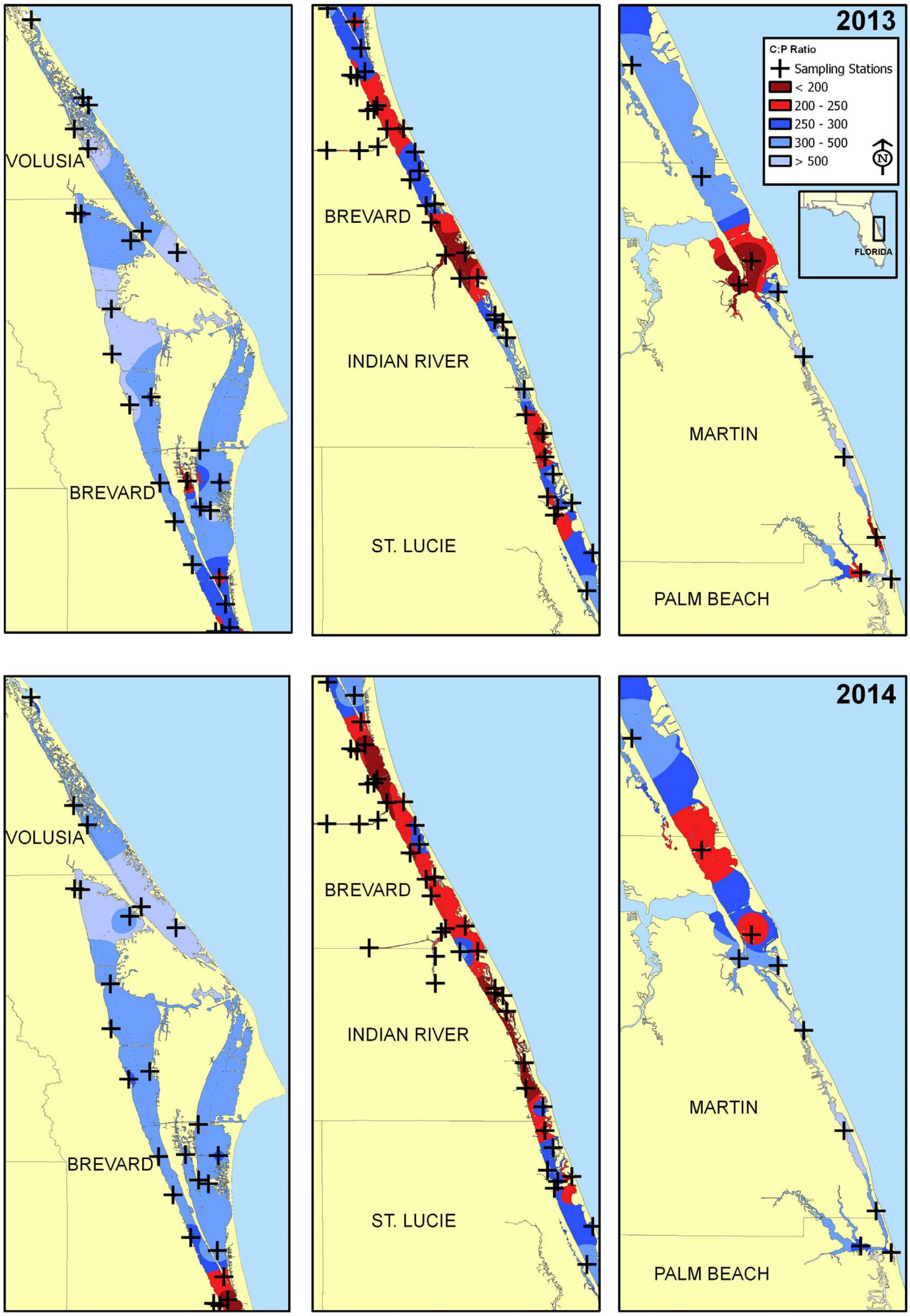


Fig. 8. Geospatial map of carbon to phosphorus (C:P) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

Like the Banana River Lagoon sub-section, algal tissue %N contents in the northern IRL increased from 2013 to 2014 (2.2% ± 0.7 versus 2.7% ± 0.9), but were variable and not significantly different between years. In both the 2013 and 2014 years, significant phosphorus-limitation in algal tissue (< 0.15% TP) and high ratios of nitrogen to phosphorus (N:P > 30, values up to 71) characterize the northern Indian River Lagoon sub-section. Phosphorus availability in algal tissue increased to the south, during both years in this sub-section (Fig. 6). Phosphorus contents are also high in algal bio-indicators adjacent to WWTPs, and at rural and agricultural sites at the northern terminus of the northern IRL.

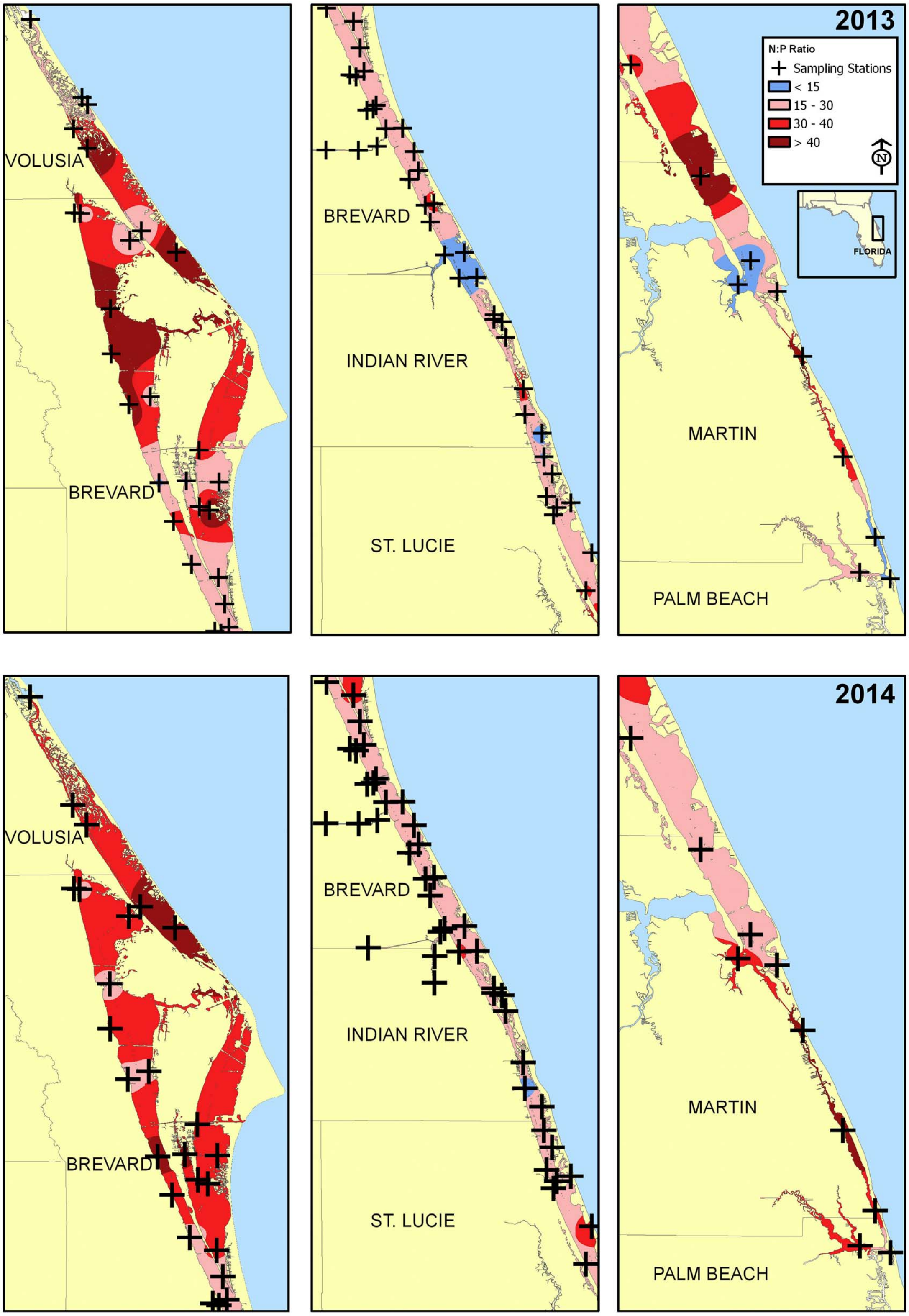
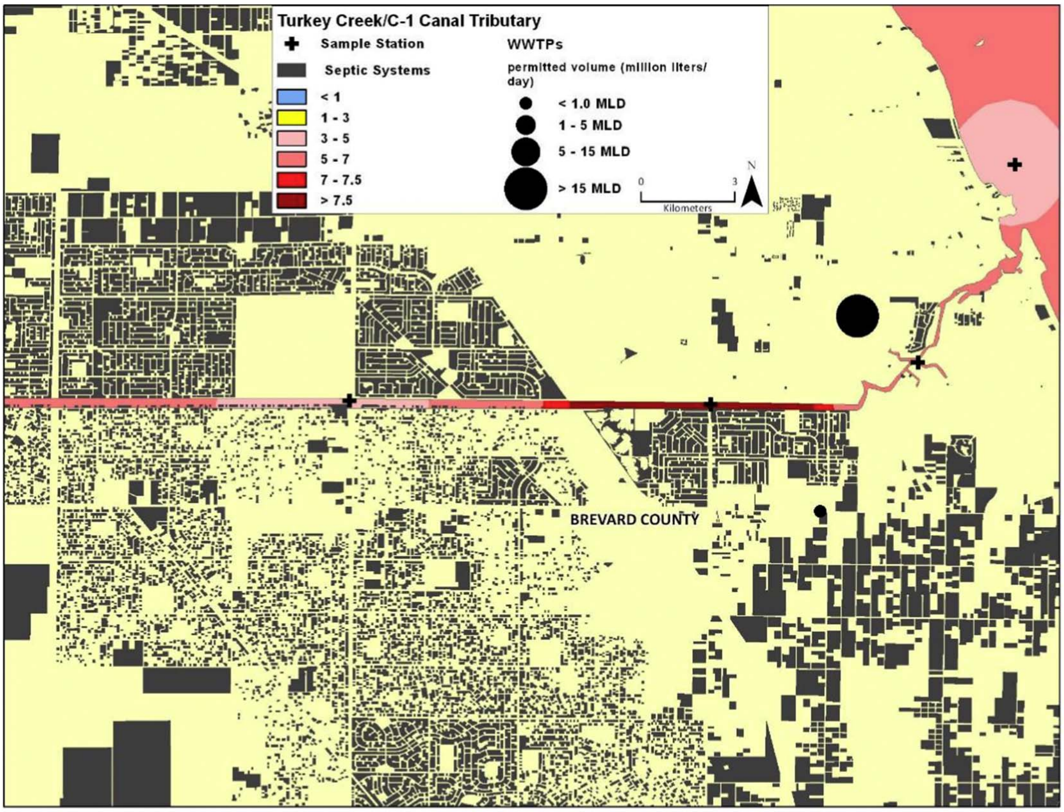


Fig. 9. Geospatial map of nitrogen to phosphorus (N:P) values from macroalgae in the Indian River Lagoon system in 2013 and 2014.

14. Central Indian River Lagoon

The heavily urbanized central IRL sub-section also had significantly higher $\delta^{15}N$ values in 2013 versus the wetter 2014 (> +7.0‰, versus +5–7‰) at sites across the urbanized upper portion sub-section (see Table 2, Fig. 4). The lower portion of the central sub-section is less urbanized (Sebastian to Vero Beach mainland and barrier island), with a larger proportion of residences on OSDS (see Table 1), versus the more urbanized Melbourne area with a higher proportion of residences in municipal WWTP service areas. The highest $\delta^{15}N$ value (+12.7‰) from the study was found at a site adjacent to the South Beaches WWTP across the Lagoon from Palm Bay in 2013, where highly nitrified (treated) wastewater effluent is irrigated on the barrier island golf course. However, values at that site were lower (+8.1‰) in 2014,



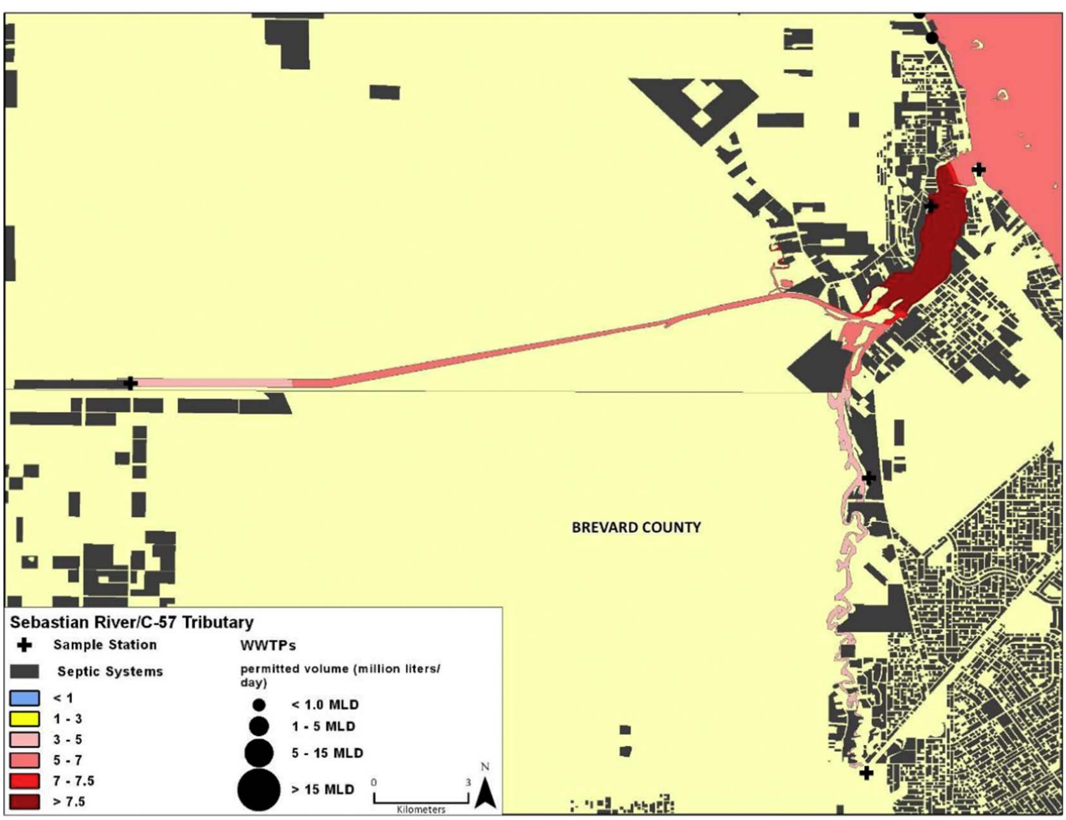


Fig. 10. Geospatial map of $\delta^{15}N$ (‰) values in macrophytes from the central IRL system Turkey.
Creek tributary (Palm Bay) and St. Sebastian River tributary (Sebastian) and residences utilizing Onsite Sewage Disposal Systems (OSDS) in black polygons in adjacent watersheds. Municipal wastewater treatment plants (WWTP) and respective treatment volumes are denoted in black spheres.

Table 2
Annual and wet season antecedent (July through September) rainfall totals (cm) and percent differences between 2013 and 2014 for NOAA-NWS meteorological stations along the IRL system, Daytona (12834), Melbourne (12838), Vero Beach (12843), St. Lucie (12893), Stuart (18) and Jupiter (84461).

	Annual			Wet season		
	2013	2014	Δ	2013	2014	Δ
Daytona	122	162	+33	48	71	+48
Melbourne	108	145	+34	35	71	+102
Vero Beach	113	137	+21	35	59	+69
St. Lucie	120	144	+20	48	58	+20
Stuart	136	129	−5	65	46	−41
Jupiter	242	229	−6	87	102	+17

Table 3
Changes in annual and wet season (July to September) antecedent rainfall totals from 2013 to 2014 adjacent to the 5 sub-sections of the IRL system. Differences in annual mean values of Δ δ^{15}N and Δ%N in macroalgae from 2013 to 2014 are listed for respective sub-sections. Significance between annual mean values from IRL subsections are denoted at $p < 0.001$ = **, $p < 0.05$ = *, and not significant = NS.

IRL sub-section	Δ Annual total	Δ Wet season total	Δδ^{15}N	Δ%N
Mosquito Lagoon	+33%	+48%	−0.2^{NS}	+0.4*
Banana River Lagoon	+34%	+102%	−3.0**	+0.3 ^{NS}
IRL- Northern	+34%	+102%	−1.8**	+0.5 ^{NS}
IRL- Central	+21%	+69%	−1.3*	+0.4*
IRL-Southern	−5%	+20%	−2.9**	−0.1 ^{NS}

when less retention time prior to “emergency discharges” resulted in less nitrification of effluents. The 2013 value from macroalgae in the adjacent surface water, representing “reclaimed water” nitrogen appears distinct from other wastewater sources detected in the IRL system. In two central IRL tributaries, Turkey Creek and Sebastian River, elevated δ^{15}N values (> +7.5‰) in macrophytes adjacent to residential areas with OSDS use are presented in Fig. 10.

Tissue nitrogen contents also increased significantly from the dryer 2013 to the wetter 2014 year (2.1% ± 0.6 versus 2.5% ± 0.5, $p < 0.05$, see Table 2), particularly in the southern portion of the subsection, near Sebastian in Indian River County. Here, %N values increased from ~1.0–2.0% to > 2.0%. Tissue phosphorus contents were high (> 0.2%) throughout the majority of this portion of the IRL in both years (Fig. 6), and were highest (> 0.3%) adjacent to the urban areas of Palm Bay (Turkey Creek), Sebastian and Vero Beach. Also, during the wetter 2014 year, nitrogen to phosphorus ratios (Fig. 9) were significantly higher than 2013 (21.3 ± 5.8 versus 18.9 ± 7.5, $p < 0.001$), indicating the significant role of nitrogen enrichment in elevating N:P ratios during a more rainy year.

15. Southern Indian River Lagoon

The southern IRL sub-section experienced a significant decrease of +2.9‰ in algal tissue δ^{15}N values from 2013 to 2014 (see Table 3). Annual rainfall totals in the southern terminus of the IRL (Jupiter) were twice the annual totals versus those in the central and northern IRL regions, and unlike to the northern IRL, the southern sub-section had similar annual and wet season rainfall totals between 2013 and 2014. The southern IRL also receives significant riverine input from Taylor Creek and St. Lucie River with adjacent municipalities and rural areas heavily reliant of OSDS' for residential wastewater treatment. During 2013, evidence of high δ^{15}N values (+7.0‰) adjacent to areas with high OSDS density near the Sewell's Point to Jensen Beach area, deceased to +4.0‰ in the 2014 year. Concentrations of algal tissue nitrogen (< 2.0%) and phosphorus (< 0.2%) were both low, and not significantly different between years. Nitrogen to phosphorus ratios in algal tissue rose from 23 to 29 in 2013 to 2014, but were not significantly higher. Some of the lowest δ^{15}N values of < +3.0‰) in the study were measured in this portion of the Lagoon where the predominant bloom species *Lyngbya majuscula* is a facultative nitrogen fixer, depending on environmental conditions.

16. Resolving external nitrogen sources into IRL system harmful algal blooms

High resolution sampling of macroalgal bio-indicators for identification and discrimination of multiple land-based sources of nitrogen using δ^{15}N has become routine in coastal environments, worldwide. For example, the technique has been used in Australia's Great Barrier Reef inshore waters (Costanzo et al., 2001), Sweden's fjords (Savage and Elmgren, 2004), reefs in Hawaii (Dailer et al., 2010), Japan (Umezawa et al., 2002), Tobago (Lapointe et al., 2010), the Bahamas (Barile and Lapointe, 2005), NE US estuaries (McClelland and Valiela, 1998), the Florida Keys (Lapointe et al., 2004), and littoral and coastal reef systems along the east coast of Florida (Barile, 2004, Lapointe et al., 2005). Along the Indian River Lagoon system, multiple anthropogenic nitrogen sources, including agricultural and urban fertilizer, atmospheric deposition, and human wastewater are recognized as significant nitrogen loading sources (Janicki Environmental and Applied Technology and Management, 2012) that may contribute to the IRL's eutrophication and harmful algal bloom (phytoplankton and macroalgae) problems.

The broad length of the IRL system (~240 km), provided the opportunity to discriminate various nitrogen sources adjacent to rural and relatively unimpacted areas, such as the southern Mosquito Lagoon which is surrounded by the federally-protected Canaveral National Seashore and Merritt Island National Wildlife Refuge. Here, δ^{15}N values were consistent with atmospheric nitrogen deposition source signatures (δ^{15}N = +1–3‰), as there are no fertilizer or human wastewater sources in the adjacent upland. Alternatively, much of the IRL system lies adjacent to heavily urbanized municipalities, many with significant residential OSDS use in low lying areas (see Table 1, Fig. 2), and municipal wastewater infrastructure with problematic performance records during wet weather conditions (FDEP, 2017). In both dryer and wetter years, the majority of the IRL system (including the 5 discrete sub-sections (ML, BR, IRL-N, IRL-C, IRL-S) all had macroalgal bio-indicator δ^{15}N values > +3‰, and in the +7 to +10‰ range for the most tidally-restricted urbanized areas, indicating that nitrogen from human sewage is the primary source supporting macroalgae bloom species across the IRL system. Further, elevated δ^{15}N values in the significant tributaries of the central IRL (see Fig. 10) with heavy reliance on residential OSDS, such as Turkey Creek (δ^{15}N mean = +6.6‰) and the St. Sebastian River (δ^{15}N mean = +6.0‰), suggest significant human wastewater advection to the downstream IRL system. These δ^{15}N results for the St. Sebastian River are corroborated by Tarnowski (2014), who also followed OSDS effluents in groundwaters to surface waters using inorganic nutrients, δ^{15}N and sucralose tracers. Alternatively, only two sites at the urbanized (and sewered): 1) Turkey Creek (δ^{15}N mean = +1.7‰) in 2013, and 2) the Vero Beach Moorings (δ^{15}N mean = < 3.0‰) indicated urban fertilizer as a primary nitrogen source. With no adjacent anthropogenic land-based sources, macroalgal bioindicators at sites in the lower portion of the Mosquito Lagoon, surrounded by the federally protected Canaveral National Seashore and Merritt Island National Wildlife Refuge consistently indicated (δ^{15}N means = +1 to 3‰) atmospheric deposition as the primary nitrogen source. Another important finding, here, is the emergence of *Lyngbya majuscula*, a facultative nitrogen-fixing blue-green macroalgae that replaced autotrophic macroalgae at the southern IRL study sites between St. Lucie and Jupiter Inlets in 2014, dropping the δ^{15}N value in this area from the 2013 mean δ^{15}N = +5.8‰ to δ^{15}N = +1.6‰ in 2014. These cyanobacteria macroalgal blooms have replaced seagrass and other macroalgal communities in similar nitrogen-saturated coastal waters, in conditions that destabilize succession of benthic macrophytes. Evidence suggests that when sensitive subtropical ecosystems become saturated with sewage nitrogen, chelators in wastewater, such as EDTA, provide competitive advantages for *Lyngbya majuscula* that replace seagrass and macroalgal communities (see Bell and Elmetri, 2007).

This finding of human wastewater as the primary land-based anthropogenic nitrogen source is consistent with a more limited spatial analysis performed by Lapointe et al. (2015) for the IRL system, which indicated a Lagoon-wide mean of $\delta^{15}N = 6.3‰$. Further, it is supported by findings of human sewage as the primary nitrogen source fueling harmful algal blooms in coastal estuaries around Florida, such as Sarasota Bay (Dillon et al., 2000), Charlotte Harbor (Lapointe and Bedford, 2007), the Florida Keys (Lapointe et al., 2004) and the St. Lucie River (Lapointe et al., 2012, 2017), a significant tributary of the southern IRL.

There is mounting evidence that human wastewater pollution is a driver of higher food web dysfunction in the Indian River Lagoon. Pathogens associated with human wastewater, such as anti-biotic resistant bacteria and viruses have been identified in IRL bottlenose dolphins (Schaefer et al., 2009). Schaefer et al. (2011) has reported that areas of the IRL with high densities of OSDS are a significant risk factor for *E. coli* contamination of the Lagoon's bottlenose dolphins. Dolphins in the IRL were six times more likely to be colonized with *E. coli* in sections of the Lagoon with > 80,000 OSDS, versus sections of the IRL with < 70,000 OSDS. This relationship is not surprising, given the direct evidence of significant human fecal contamination of ground and surface waters adjacent OSDS along the IRL (Lapointe et al., 2012, 2017). Further, sewage-driven blooms of phytoplankton and macroalgae were reported to produce toxins and resulted toxin accumulation and die-offs of marine mammals (manatee) that have consumed these bloom species in the central Lagoon area (Lapointe and Herren, 2015).

17. Septic tanks deliver more nitrogen and support blooms in the wet season

The physical role of hydrodynamics, nutrient biogeochemistry and physiology of algae on spatial and temporal scales may ultimately dictate which sources are most significant in supporting harmful blooms of algae and eutrophication. Specifically, dry to wet season shifts in rainfall, changes in stormwater hydroperiod, hydraulic pressure on shallow groundwaters and subsequent residence time in groundwater mobilization to estuaries can change both groundwater and receiving water $\delta^{15}N$. For example, highly nitrified septic tank effluent in adjacent groundwater during dry seasons are commonly > +10‰ (Lapointe and Krupa, 1995a). However, during wet seasons with less retention time, ammonium-N predominates as an effluent N-source, where septic tank effluent $\delta^{15}N$ is reported to be +4 to +5‰ (see Lapointe and Krupa, 1995a, Hinkle et al., 2008, Katz et al., 2010, and Bicki and Brown, 1990). The lowering of $\delta^{15}N$ values in coastal waterbodies heavily reliant on OSDS during the wet season has been reported in tributaries of the IRL system by Lapointe et al. (2017) for the St. Lucie River (tributary of the southern IRL) system, and Tarnowski (2014) for the Sebastian River (tributary of the central IRL). But also for areas downstream of significant agricultural areas Lapointe and Bedford (2007) for Lee County, and Lapointe et al. (2004) in the Florida Bay. In larger watersheds with agriculture and residential areas on municipal wastewater treatment, multiple sources, including fertilizers and atmospheric deposition resulting from wet season surface runoff may also contribute to mixing of sources (see McClelland and Valiela, 1998) resulting in lowering of $\delta^{15}N$ values in macrophytes in downstream surface waters. But, in Florida estuaries, these source signals (< +3‰) appear to be discriminated from higher $\delta^{15}N$ (~+8‰) signals adjacent to wastewater treatment plants or tributaries and canals with high OSDS densities, such as in Sarasota Bay (see Dillon et al., 2000), and the Florida Keys (Dillon et al., 2000, Lapointe et al., 2004).

Sewage nitrogen has been identified as a significant contributor to eutrophication in many coastal ecosystems throughout Florida, including the Indian River Lagoon (see Lapointe et al., 2015). The fine-scale spatial evidence provided here using biochemical analyses of estuarine macroalgae and comparing known human wastewater pollution sources suggests that sewage nitrogen is a significant source supporting macroalgal biomass across most of the 240 km long Indian River Lagoon system.

18. Nitrogen saturation and harmful algal blooms in the IRL

The results of this study support previous findings of elevated nutrient enrichment of the IRL system during wet season conditions, where increases in water column nutrient concentrations drive initiation and expansion of harmful algal blooms (see Lapointe et al., 2015). Here, higher % nitrogen tissue contents in the Indian River Lagoon system (see Table 3., Fig. 5) provides evidence that benthic macroalgal communities are undergoing "luxury storage" of excess dissolved inorganic nitrogen in many sub-sections of the IRL, beyond what is necessary to support photosynthesis. In west coast US estuaries, Cohen and Fong (2006) reported higher % nitrogen concentrations in macroalgae adjacent to significant loading sources. Elevated nitrogen concentrations in wet summer and El Nino wet winter seasons support expansion of macroalgal blooms (Lapointe et al., 2015). With advanced eutrophication of the IRL system, toxic phytoplankton, such as the dinoflagellate *Pyrodinium bahamense* var. *bahamense* have been reported where "excess bio-availability of nutrients" are common in the northern and central IRL system (Phlips et al., 2004). This species has been reported to be a toxin producer, with saxitoxin being isolated from *Pyrodinium bahamense* var. *bahamense*, and movement of saxitoxin into food web components, such as finfish (Landsberg et al., 2006).

Replacement of seagrass communities with macroalgal blooms has occurred in the IRL system since the late 1980's (see Barile and Lapointe, 1999, Hall et al., 2001). Ultimately, expansive macroalgal die-off, fish kills and die-offs of marine mammals coincided with continued expansion of toxic phytoplankton blooms over the past decades (see SJRWMD, 2012).

More recently, the IRL system brown tide *Aureoumbra lagunensis* bloom organism, recognized to preferentially uptake reduced forms of nitrogen (> 90% as NH_4^+ and urea), underwent significant bloom expansion in the 2012 and 2013 summer wet seasons in the Mosquito and northern Indian River Lagoon sub-sections (Kang et al., 2015), and most recently in the historically wet El Nino winter of 2016. In the southern Indian River Lagoon, the primary tributary, the St. Lucie Estuary, significant blooms of the cyanobacteria *Microcystis aeruginosa*, including toxic microcystin-producing strains, bloomed in urban areas where sewage nitrogen and phosphorus were at the highest concentrations of NH_4^+ and reactive phosphorus (see Lapointe et al., 2017). Lapointe et al.'s (2017) conclusion that the toxic strain of this *Microsystis* bloom is supported by urban human wastewater is supported by water column $\delta^{15}N$- NH_4^+ values that matched tissue $\delta^{15}N$ values in *Microcystis* blooms. However, despite the need for elevated concentrations of reactive nitrogen, both of these blooms occur in portions of the IRL system where high N:P ratios occur (see Fig. 9). These two blooms are case studies which indicate that as nitrogen saturation of estuarine environments occurs and water column N:P ratios increase beyond the Redfield Ratio (i.e. > 15:1 of N to P), phytoplankton bloom species have developed physiological adaptations to: 1) subvert previously recognized stoichiometric limitations, where phosphorus may limit bloom formation (see Lapointe et al., 2017, and Kang et al., 2015), and 2) increase toxicity of bloom species in the IRL system and its tributaries e.g. *Microcystis* (see Lapointe et al., 2017) where N:P ratios are high. Likewise, the highest reported bloom cell densities of the saxitoxin-producing *Pyrodinium bahamense* var. *bahamense* were from the northern IRL where N:P ratios (> 25) are most elevated (Phlips et al., 2006).

19. Are IRL system HABs supported by external nitrogen or internally-derived "muck" nitrogen?

Many IRL system resource managers have identified "muck removal" as a primary strategy for mitigating nitrogen and phosphorus sources that support HABs (Brevard County Natural Resources Department, 2017). This is despite any published evidence that NH_4^+ fluxes from benthic muck (~10% organic matter) deposits either: 1) are spatially relevant as a source to the significant northern IRL or Mosquito Lagoon HABs, or 2) are directly supporting HABs as a demonstrable nitrogen source in the IRL system (see Trefry, 2013). Alternatively, for HAB species in the IRL system, the evidence suggests that blooms initiate and expand during or in response to episodic seasonal or climatologically-episodic rainfall events. Phlips et al. (2006) noted that the saxitoxin producing *Pyrodinium bahamense* var. *bahamense* blooms that occurred periodically over 7 years of study in the northern and central IRL, showed a response to long-term climatic cycles and stochastic rainfall events. In contrast, biomass of this bloom species was low in drought periods, but dramatically elevated during the high rainfall wet seasons of 2001 to 2004. Similarly, large blooms of this species occurred in Tampa Bay in the summer of 2002, where there are no reports that internally-derived nitrogen from muck deposits are a significant loading source versus external nitrogen loading in the wet season. For the brown tide *Aureoumbra lagunensis* HABs of 2012 and 2013, Kang et al. (2015) reported that high $\delta^{15}N$ values (~+5‰) for particulate organic nitrogen (PON) in the IRL system are indicative of a sewage nitrogen source from OSDS in the northern IRL and Mosquito Lagoon areas. However, once initiated in the wet season, $\delta^{15}N$ values of PON during *Aureoumbra lagunensis* blooms decreased to (~+3‰), as the blooms preferentially uptake more NH_4^+ from pelagic nitrogen recycling resulting from microbial remineralization, thereby reducing $\delta^{15}N$ values. Also, these authors suggest that higher water column NO_3^- in non-bloom periods likely results in higher PON $\delta^{15}N$ values during these conditions. Lapointe et al. (2015, 2017) both reported that macroalgal blooms in the IRL system and its tributaries expand with wet season fluxes of human sewage-derived inorganic nitrogen from OSDS' into groundwaters, surface waters and into macroalgal bloom tissue. The findings from these case studies are consistent with this study, where significant wet season rainfall drives external nitrogen loadings of human wastewater from upland watersheds, as the primary source supporting HABs in the IRL system.

20. Policy implications

By 1990, the Indian River Lagoon, Tampa Bay, and Sarasota Bay were all designated as EPA-National Estuary Programs. On the west coast of Florida, mitigation of human wastewater loading was a primary strategy for restoration of both Tampa Bay and Sarasota Bay. For Tampa Bay, in the early 1980's, conversion of 9 WWTPs to Advanced Wastewater Treatment (AWT), resulted in 90% mitigation of wastewater nitrogen and a ~60% reduction of total N load (Greening and Janicki, 2006). Seagrasses have since recovered to percent cover levels not seen since the 1950's (Greening et al., 2014), and is a globally-recognized case study of a restored estuary. In Sarasota Bay, conversion of municipal WWTPs to AWT, likewise, reduced nitrogen loads to the estuary by 46%. However, non-point nitrogen loading, including nitrogen loads from 14,000 residential OSDS in the Phillipe Creek tributary, were identified as a significant pollution source in 1997 (Burden et al., 2003). Subsequently, an aggressive septic-to-sewer conversion program, mandated by Sarasota County, in the Phillipe Creek tributary, mitigated over 11 million liters per day of baseflow associated with septic tank effluent into the Creek and into Sarasota Bay. Sarasota Bay's seagrass coverage in 2014 exceeded coverage in 1950 by 30% (SWFWMD, 2015), and the Bay meets State of Florida Impaired Water's List Numeric Nutrient Criteria for nitrogen and phosphorus. Similarly, the State of Florida has completed a $1 billion project to convert 76,000 OSDS to AWT municipal wastewater treatment in the Florida Keys, following evidence of wastewater impacts resulting in the demise of the third largest coral reef tract on earth.

Alternatively, for the Indian River Lagoon, legislation of the State of Florida's "Indian River Lagoon Act" in 1990, required the mitigation of municipal wastewater discharges to the IRL system. However, very few municipal WWTPs along the IRL were converted to high nutrient removal AWT. Although routine surface water discharge is no longer permitted for most WWTPs adjacent to the Lagoon, wet weather emergency discharge is permitted for 90 days per year. Consequently, significant wet weather discharges of nearly 15 million liters were reported for the Palm Bay- Turkey Creek WWTP during the 2014 sample period (FDEP, 2017). These permitted Emergency discharges are not always reported in official State of Florida spill reports, and reporting is not required by the federal EPA. Thacker (2004) reported that, across the US, over 3200 billion liters of effluent are discharged annually into adjacent surface waters in emergency conditions. The American Society of Civil Engineers (2016) reported that Florida has $18.4 billion dollars (US) in wastewater infrastructure upgrade costs necessary to accomodate population growth and protect adjacent water quality. In Brevard County, which borders ~40% of the IRL system, there is a $159 million deficit in county wastewater treatment infrastructure (see Brevard County Utilities Services, 2013), when including the major cities in that county, that cost estimate is likely double.

In the southern IRL's Loxahatchee River tributary, Lapointe and Krupa (1995a and 1995b) reported evidence of the couplings of residential OSDS, contamination of groundwaters, downgradient surface waters, and decline of adjacent seagrass ecosystems. This evidence was used in a State of Florida administrative hearing to compel mandatory septic to sewer conversion in the town of Tequesta and Jupiter. Although, studies with similar results have been produced in Martin (Lapointe et al., 2012, 2017) and Indian River Counties (Tarnowski, 2014), the Indian River Lagoon National Estuary Program, and many participating municipalities and county governments have been slow to advocate similar wastewater treatment infrastructure upgrades (i.e. in Sarasota Bay NEP, and Florida Keys) to mitigate coastal water quality decline and habitat impacts.

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Appendix A. Supplementary data

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