Research Article

Remote sensing of harmful algal blooms in the Indian River Lagoon and connected waterways in Brevard County

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Abstract With an increasing frequency of Harmful Algal Bloom (HAB) occurrences in the Indian River Lagoon (IRL) over the last decade, the identification of potential HAB triggers and hotspots of activity is vital to the Lagoon's restoration efforts. From 2015 to 2021 there were three major HAB events identified through surface water monitoring: 2016, 2018, and 2020. The use of remote sensing technologies is a cost-effective and encompassing approach for providing rapid identification of HAB formation, the HAB stages, and identification of hotspots of HAB occurrences. The Normalized Difference Chlorophyll Index (NDCI) was applied to the European Space Agency Sentinel 2 and Sentinel 3 satellite data over the IRL. The S2 NDCI algorithm had an R^2 of 0.81 and root mean square error (RMSE) of 14.14 micrograms per liter (μg/L) for the calibration dataset of 84 points ranging from 1.3 to 154 μg/L ChlA. The S3 NDCI algorithm had an R^2 of 0.92 and an RMSE of 10.01 μg/L for the calibration dataset of 153 points. Ranging from 0.4 to 187 μg/L. A Time Series Clustering and Emerging Hotspot analysis was performed on the Brevard County portion of the IRL utilizing the S2 NDCI estimated Chlorophyll A (ChlA) concentrations. A total of six significantly different clusters of ChlA variation were identified in the IRL. Of these six, the North-North IRL and Central-North IRL were the only two clusters to have statistically significant increasing trends in ChlA. The emerging Hotspot Analysis identified the Central-North IRL and North BRL as two segments of the IRL which appear to have the most frequent and intense bloom activity. The rapid identification and analysis of HAB activity through the Sentinel satellites can provide the managers and stakeholders of the IRL with valuable warning and planning information.

1.0 Introduction

With an increasing frequency of harmful algal blooms (HAB) occurrences in the Indian River Lagoon (IRL) over the last decade, the identification of potential HAB triggers and hotspots of activity is vital to the Brevard County efforts to manage the watershed. For decades, the integrity of the IRL has been negatively impacted by the persistent occurrence of HABs that lead to low dissolved oxygen, limited light availability as well as subsequent losses of important flora and fauna (Lapointe et al., 2020). The most prevalent HAB species reported throughout the IRL over the last decade by biomass was *Aureoumbra lagunensis*, while *Pyrodinium bahamense*, and *Pseudo-nitzchia* spp. were also frequently observed at bloom levels (Phlips et al., 2021; Lopez et al., 2021). From 1997 to 2010, *P. bahamense* was the dominate algal species present, concentrating in the North IRL (NIRL) near Titusville (Phlips

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et al., 2021); however, after the 2011 "superbloom" event that persisted for 7 months, there was a major loss of seagrass (>40%) and a drastic shift in species composition and bloom intensity was reported with *A. lagunensis* as the new dominant species of algae (Morris et al 2021, Phlips et al., 2021). Additionally, picocyanobacteria and nanoplanktonic species played a dominant role in the overall regime shift in 2011, as it has been hypothesized that increased concentrations of ammonium and organic forms of nitrogen and phosphorus provide an advantage for brown tide and other pico-nanoplanktonic species competing for nutrients (Phlips et al., 2021). Knowledge of phytoplankton diversity and density was determined by an intensive surface water sampling effort, however due to logistical and funding constraints, spatial and temporal gaps exist in these datasets.

The use of remote sensing technologies is a cost-effective and encompassing approach for providing rapid identification of HAB formation, the HAB intensity, and identification of hotspots of HAB occurrences. Airborne and satellite remote sensing provides a spatially continuous landscape estimate of conditions and with satellite remote sensing, daily or weekly measurements. This high resolution spatial and temporal data can provide information that monthly, widely spaced surface water grabs may miss. Different proxies have been utilized for detection of HABs, the most common being chlorophyll-a (ChlA, Khan et al., 2021). Of the pigments active in photosynthesis, ChlA has the strongest influence on the reflected spectral signal of water (Maxwell & Johnson 2000). ChlA preferentially absorbs energy in the blue and red spectra then reflects green, resulting in the relatively high reflectance in the green spectrum (Morel & Prieur 1977; Tedetti et al., 2011). The ChlA pigment also has a strong reflectance in the Near Infra Red (NIR), specifically around 708 nm (Gitelson et al., 1996; Gower et al. 2008). The estimation of ChlA variation is commonly performed using band ratios, which leverage the difference between the ChlA reflectance features, are resistant to confounding factors in the water column and allow the comparison between scenes of data collected at different times (Cabellero et al., 2021, Ogashawara 2013, Rodriguez et al., 2020). One of the more common ratios is the Normalized Difference Chlorophyll Index (NDCI) which was developed for European Space Agency (ESA) Medium Resolution Imaging Spectrometer (MERIS) and is the ratio of the difference over the sum between the 708 nm and 665 nm spectra (Red edge) (Mishra & Mishra 2012, Rodríguez-Benito et al 2020).

The ESA continued the legacy of MERIS which was successfully used to map the 2011 "superbloom" event (Kamerosky et al 2015) with the Sentinel-2 (S2) and Sentinel-3 (S3) satellites. The S2 Multi Spectral Imager (MSI) launched in 2015 aboard S2A and 2017 aboard S2B, along with the S3 Ocean and Land Color Instrument (OLCI) which launched in 2016 aboard S3A and 2018 aboard S3B. The S2 and S3 satellites provide high spatial, spectral, and temporal resolution as well as having a proven record in published literature for the evaluation of HABs (Caballero et al 2020, Kravitz et al 2020). The MSI has 10 bands in the visible to NIR spectrum, with a spatial resolution between 10-60 m, and a revisit time less than 2 days over Florida. The OLCI has 21 bands in the visible to NIR spectrum, a spatial resolution of 300 m, and has a revisit time less than 2 days over Florida.

The Sentinel satellites when paired with the monthly water quality monitoring data collected by the St Johns River Water Management District (SJRWMD) can accurately estimate the variability of IRL HABs in space and time. These identified patterns can then be further examined to determine their potential causes and effects on the aquatic ecosystems of the IRL. This paper examined the S2 and S3 data over the IRL to identify the continued applicability of the NDCI ratio to characterizing HABs in the IRL. The ChlA estimated via IRL Sentinel imagery from 2015-2021 was then used to perform a spatiotemporal analysis of ChlA patterns. The objective of this study is to determine the applicability of spatiotemporal analysis of estimated ChlA concentrations within the IRL following the specific hypotheses below:

Hypothesis 1: Within the IRL, the 708 nm red edge reflectance peak can explain most variation in ChlA concentrations.
Hypothesis 2: HAB intensity, scale, and duration has been increasing throughout the IRL from 2015 to 2021.

2.0 Methods

2.1 Site Description. The IRL is an Estuary of National Significance within the EPA National Estuary Program with more than 1 million residents in its increasingly developed watershed (US Census 2021). The IRL is located along 250 km (155 miles) of the Florida east coast with over 750 square km (290 square miles) of surface water, between multiple jurisdictions with a range of surrounding land cover types and benthic environments. The project is focused on the basins of the IRL within Brevard County (Figure 1), which includes Mosquito Lagoon (ML), North IRL (NIRL), Banana River (BR), and Central IRL (CIRL).

2.2 Surface Water Quality Monitoring. *In-situ* water quality data from the St. Johns River Water Management District's (SJRWMD) monthly surface water quality grab sites (Figure 1) were collected to calibrate Sentinel imagery. Available corrected ChlA was obtained for the stations from January 2015 to August 2021 using the SJRWMD Environmental Data Retrieval Tool (http://webapub.sjrwmd.com/agws10/edqt/). For surface water quality sites IRLB04, IRLML02, IRLI13, and IRCMTITUS01, continuous monitoring (CM) data are collected and regular monthly discrete samples are collected later in the month when the station is serviced. A total of 23 monthly sampling stations were used in this study.

Corrected ChlA is determined by removing the influence of pheophytin as per FDEP SOP DEP-SAS-002/10. Pheophytin is a degradation product of ChlA, which can result in erroneously high ChlA measurements. Between July and December 2020 there was a J qualifier code (Estimated Data) assigned to the IRL corrected ChlA values. The uncorrected ChlA concentrations were used in place of these values. In October 2017, the ChlA measurements at IRLI10 through IRLI21 were excluded from analysis due to concerns over data accuracy. In September 2019, the ChlA measurement at IRLIRCMTITUS01 was excluded from analysis due to concerns over data accuracy.

2.3 Sentinel 2 and 3 Imagery Acquisition and Processing. Satellite remote sensing data from Sentinel-2 (S2) and 3 (S3) satellites were acquired for their period of record starting in 2015. A total of 142 S2 and 536 S3 scenes of clear or low cloud cover were obtained. S2 and S3 imagery was downloaded through the ESA's Copernicus Open Access Hub (https://scihub.copernicus.eu/) and processed to L1C Top of Atmosphere (ToA) Reflectance and a cloud mask generated using the SentiNel Application Platform (SNAP). No atmospheric correction was applied to the imagery as these methods are likely to disrupt the red edge bands of the selected band indices. The downloaded imagery was processed and stored as a multidimensional raster dataset in ESRI ArcPro 2.9. The spectral values of pixels that were within 24 hours of the SJRWMD surface water collection were extracted through the ArcPro Multidimensional Sampling tool and stored in a Microsoft Office Excel Workbook.

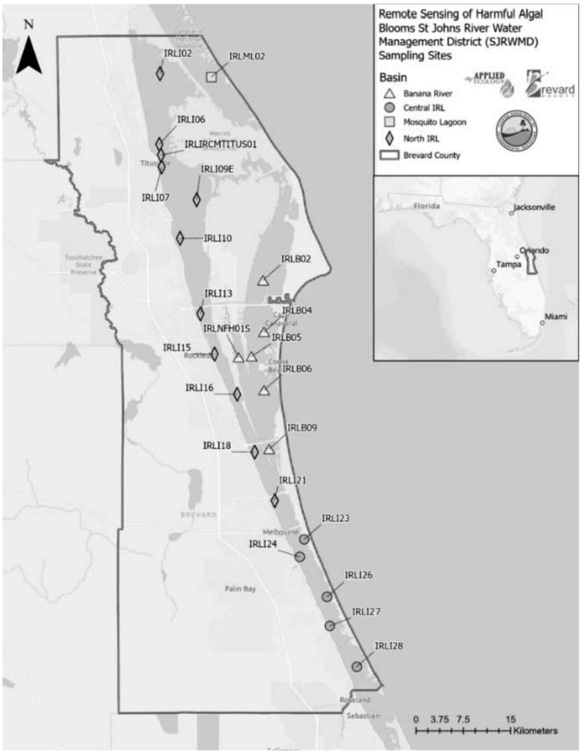

Figure 1. Study area boundary with the St Johns River Water Management District's (SJRWMD) monthly estuary surface water sampling sites identified by basin.

2.4 Statistical Analysis. 2.4.1 Model Calibration and Assessment—Analysis and calibration of the water quality and Sentinel spectral data were performed using the Microsoft Excel extension XLStat (2021.2.1). Polynomial correlations were utilized to relate the measured water quality data with the band index value and create a calibration equation. Ten percent of the available paired water quality and Sentinel data was randomly selected to be used to create a model validation dataset. Each algorithm was evaluated on its strength of correlation, Root Mean Square Error (RMSE), Bias, and the Nash–Sutcliffe model efficiency coefficient (Nash). For these equations, n is the dataset size, M is measured concentrations, $\overline{M}$ is the mean of all measured concentrations, and Est is the model estimated concentrations. The RMSE is a measure of the overall variation of the measured data from the estimation model. The Bias measure determines if the estimation model tends to underestimate (values closer to -1), overestimate (values closer to 1), or neither (values at 0). The Nash value qualifies how accurate the estimation model is by determining how well the estimated values relate the overall mean of the measured concentrations. A Nash value of 0 would mean that the estimated values are not representative of the measured values while a value of 1 would mean that there is no difference in the overall estimated and measured datasets.

Equation 1:

$$RMSE = \sqrt{\frac{1}{n}\sum_{i=1}^{n}(Est_i - M_i)^2}$$

Equation 2:

$$Bias = \frac{1}{n}\sum_{i=1}^{n}(Est_i - M_i)$$

Equation 3:

$$Nash = 1 - \frac{\sum_{i=1}^{n} (M_i - Est_i)^2}{\sum_{i=1}^{n} (M_i - \overline{M}_i)^2}$$

2.4.2 Spatiotemporal Analysis—The ESRI ArcPro Space Time Pattern Mining Toolbox was utilized to determine the ChlA trends in both space and time across the IRL as estimated by Sentinel-2. The Sentinel-2 data was used for this analysis due to the higher spatial resolution of the data which allowed for better identification and removal of confounding factors including emergent vegetation, shorelines, and bridges. The estimated ChlA values were then reclassified based on concentration ranges to improve performance by transforming the raster dataset from 32 to 8-bit rasters (i.e., 0-20 μg/L Chla to 0, 20-40 μg/L Chla to 1, 40-60 μg/L Chla to 2, 60-80 μg/L Chla to 3, 80-100 μg/L Chla to 4, >100 μg/L Chla to 5). The filtered dataset was then resampled from 20 m to 300 m due to memory limitations of the analysis tools and to reduce the impact of speckling on the analysis.

The Time Series Clustering tool was used to determine which areas of the IRL tend to increase or decrease together, delineate the spatial range of these significant clusters of the trends, and then use the Mann-Kendall Trend test to identify the significance of the trend of the cluster. The Emerging Hot Spot tools applies the Getis-Ord Gi* statistic to determine where in the IRL significant clusters of hot (high ChlA values) or cold (low ChlA values) are located and if there is a significant trend present as determined by the Mann-Kendall trend test.

3.0 Results and Data Analysis

3.1 Sentinel Calibration. The NDCI algorithm was evaluated for S2 and S3 which leverages the 708 nm ChlA reflectance band on both platforms. The S2 calibration dataset's highest ChlA value was 154 μg/L, lowest was 1.3 μg/L, and the median value 24 μg/L. This calibration dataset contained a total of 84 points with 6 points for Mosquito Lagoon, 27 for NIRL, 34 for Banana River, and 17 for CIRL. The S3 calibration dataset's highest ChlA value was 187 μg/L, lowest was 0.4 μg/L, and the median value 11.0 μg/L. This calibration dataset contained a total of 153 points with 17 points for Mosquito Lagoon, 66 for NIRL, 45 for Banana River, and 25 for CIRL. These calibration datasets are representative of the measured ChlA concentrations in the IRL.

The S2 NDCI algorithm had an R^2 of 0.81, RMSE of 14.14 μg/L for the calibration dataset, RMSE of 10.63 μg/L for the validation dataset, a Bias of 0.00 μg/L, and a Nash criterion of 0.81. The NDCI algorithm also did not have any clear outliers and as determined by the bias statistic, did not appear to consistently over- or under-estimate ChlA (Figure 2). The S3 NDCI algorithm had a R^2 of 0.92, RMSE of 10.01 μg/L for the calibration dataset, RMSE of 7.90 μg/L for the validation dataset, a Bias of -0.03 μg/L, and a Nash criterion of 0.92. The NDCI algorithm did not have any clear outliers ChlA.

3.2 Harmful Algal Bloom Spatiotemporal Analysis. 3.2.1 Satellite Estimated Chlorophyll A For 2020 Bloom Event—The most recent major bloom event observed in the SJRWMD monitoring data had started in the NIRL basin in August 2020 and ended by January 2021. Elevated bloom intensities were identified by S2 in the Central-North IRL starting in August 2020, with high bloom conditions spanning the entire NIRL by September 2020, and then in the North BR by October 2020 (Figure 3).

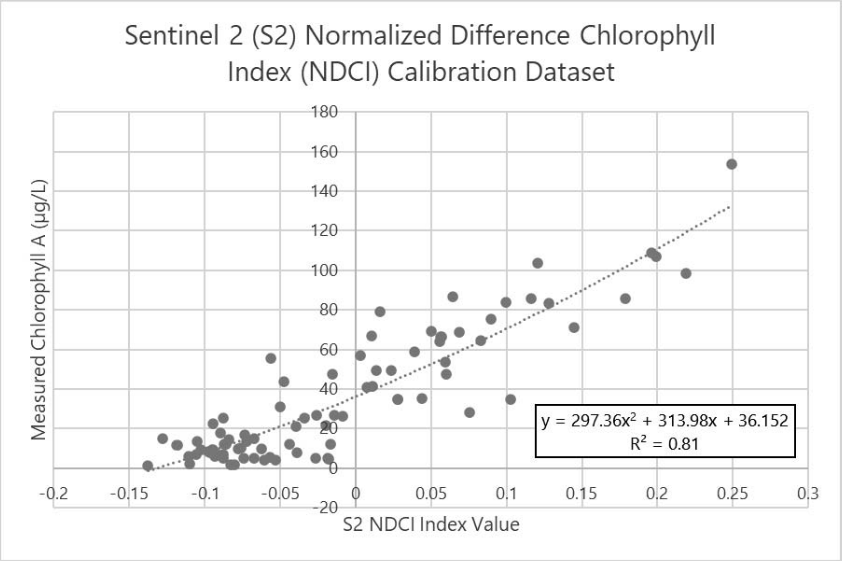

Figure 2. Normalized Difference Chlorophyll Index (NDCI) Calibration dataset for the Indian River Lagoon.

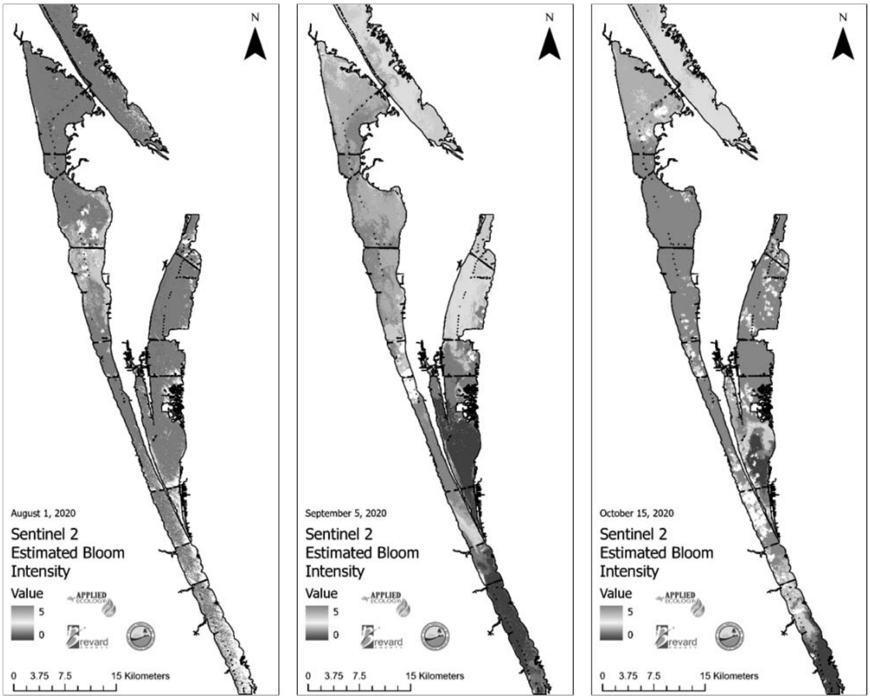

Figure 3. Sentinel 2 (S2) estimated bloom intensity for the Indian River Lagoon from August 1, 2020, to October 15, 2020.

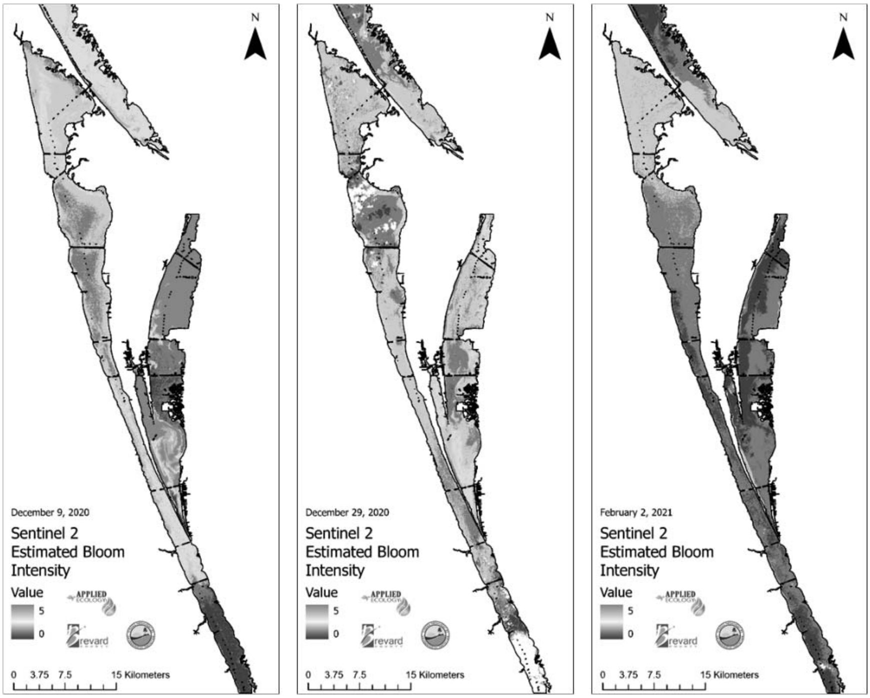

Figure 4. Sentinel 2 (S2) estimated bloom intensity for the Indian River Lagoon from December 9, 2020, to February 2, 2021.

The southern BR did not reach high bloom intensities until early December 2020 (Figure 4). From October 2020 to the end of December 2020 the blooms across the IRL began their decline, with the IRL returning to low intensities by the start of February 2021. The blooms in the North and South BR had largely declined from the start to end of December 2020, with the central portion of the BR declining first. Of particular interest is the scene from December 9th where in the southern BR there appears to have been a period of wind driven variation in the spatial distribution of the bloom.

3.2.2 Time Series Clustering Analysis—A time series clustering analysis was performed on the Brevard County portion of the IRL utilizing the S2 NDCI estimated ChlA concentrations. For the analysis, a total of 6 clusters were selected based on the optimization recommendations from the ESRI Time Series Clustering tool.

The resulting clusters of similar patterns of change in estimated ChlA concentrations mostly coincided with the SJRWMD basin delineations (Figure 5). Mosquito Lagoon and the northernmost segment of the NIRL are hydrologically connected through the Haulover Canal, suggesting that the movement of water through the canal may also act to spread a bloom. The remainder of the NIRL basin is split between Central-North and South-North clusters. The Central-North cluster

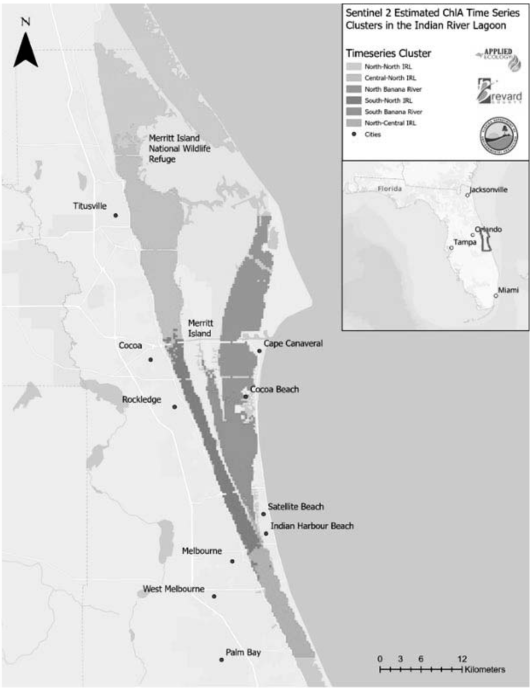

Figure 5. Results of the space time cluster analysis of the Sentinel 2 estimated bloom intensity across the Indian River Lagoon (IRL). Each cluster represents the areas of the IRL that have similar trends of change in bloom intensity.

includes, on the west shore the City of Titusville to the north and the City of Cocoa at the south, then on the east shore, the urbanized areas of central Merritt Island and natural areas of north Merritt Island. The North-North and Central-North IRL were the only two clusters to have statistically significant ($p<0.10$) increasing trends in estimated ChlA (Table 2). The South-North IRL cluster had an increasing but a marginally not significant trend ($p = 0.13$). The South-North cluster has on its west shore the City of Cocoa at the north and the City of Melbourne to the south, along with the urbanized area of South Merritt Island.

Banana River is divided into a north and south cluster, with the dividing line at City of Cocoa Beach. The North BR cluster on its west shore has the Kennedy

Table 1. Sentinel 2 (S2) Multi Spectral Imager (MSI) and Sentinel 3 (S3) Ocean and Land Color Instrument (OLCI) Normalized Difference Chlorophyll Index (NDCI) Indian River Lagoon calibration equation and statistics.

Sensor	Equation	R^2	RMSE (Calibration)	RMSE (Validation)	Bias	Nash
S2 MSI	$y = 297.36x^2 + 313.98x + 36.152$	0.81	14.14	10.63	0	0.81
S3 OLCI	$y = 437.07x^2 + 348.98x + 33.928$	0.92	10.01	7.9	-0.03	0.92

Table 2. Time series trend direction, Mann-Kendall trend statistic and p-value for the identified Indian River Lagoon (IRL) clusters.

Time Series Cluster Name	Trend Direction	Mann-Kendall Statistic	p-value
North-North IRL	Increasing	1.76	0.08
Central-North IRL	Increasing	1.94	0.05
South-North IRL	Not Significant	1.51	0.13
North BR	Not Significant	0.98	0.33
South BR	Not Significant	1.39	0.16
North-Central IRL	Not Significant	1.59	0.11

Space Center and surrounding natural areas, while the east shore has Cape Canaveral Space Force Station, the Port Canaveral lock, and the north half of the City of Cocoa Beach. The South BR has the urbanized section of South Merritt Island on its west coast and City of Cocoa Beach with Patrick Space Force Base on its east shore. Lastly, the North-Central IRL cluster runs from Melbourne to Malabar. While all three of these clusters do not have significant trends in estimated ChlA, the South BR and North-Central IRL both have positive Mann-Kendall statistics and marginally non-significant trends.

3.2.3 Emerging Hotspot Analysis—An Emerging Hotspot Analysis was performed on the Brevard County portion of the IRL utilizing the S2 NDCI estimated ChlA concentrations. The purpose of this test was to determine where in the IRL significant areas of high and low ChlA concentrations are located, then determine if these areas increased or decreased from 2015 to 2021 (Figure 6). Three patterns were detected in the IRL: Oscillating Hot Spots (OHS), Oscillating Cold Spots (OCS), and Intensifying Cold Spots (ICS).

An OHS is defined as group of cells which over time have increasing ChlA concentrations, however there is a non-random, equal distribution of the occurrence of low estimated ChlA over the entire timeseries. The OHS observed in the IRL are likely to be locations with abundant SAV coverage, as the ChlA in the SAV will also reflect and thus overestimate blooms across the entire time series with more photosynthesis in the summer and less in the winter. An OCS is defined as a group of cells that also have a positive trend in ChlA concentrations but with most of the time steps as low ChlA relative to neighboring areas, and a non-random, equal distribution of the occurrence of high estimated ChlA over the entire timeseries. An CS has a significant negative trend in ChlA concentrations over the time series and more periods of low ChlA concentrations occur over the time series.

A location classified as No Pattern Detected (NPD) has a statistically determined random distribution of high and low ChlA concentrations across its timeseries, however there is still a significant increasing trend in ChlA concentrations. There were no locations identified as New Hot Spots which are locations with few occurrences of hot spots at the start of the time series and that significantly increase over time, or Intensifying Hot Spots which are locations that had significantly high occurrences of hot spots and have increasing concentrations of ChlA over the time period.

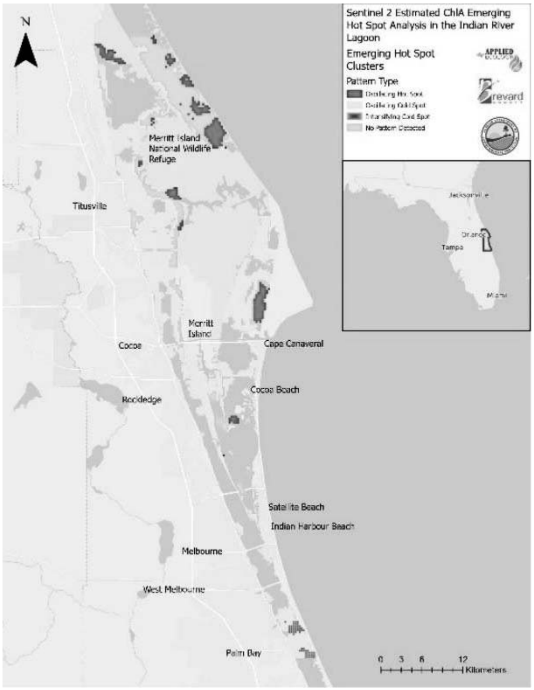

Figure 6. Emerging hot spot analysis of the Sentinel 2 estimated Chlorophyll A across the Indian River Lagoon from 2015-2021

The majority of Southern Mosquito Lagoon (SML) was classified as NPD (72%) with 5 distinct clusters of OHS (Table 3). These OHS locations were hot spots on average 41.9% of the time series and cold spots 33.2%, compared to the 27.6% hot spots and 48.4% cold spots of the NPD. The OHS locations are likely to be shallow water with SAV benthic conditions, where there are consistently high estimated ChlA with an annual reduction during the winter. The NPD area are likely to be deeper waters and the larger percent of cold spots would suggest the sporadic occurrence of blooms. There is a cluster of OCS proximate to the Haulover Canal connection between the Mosquito Lagoon and NIRL, with on average 13.2% hot spots and 66.9% cold spots. The OCS area next to Haulover Canal may be the result of the connection to the NIRL and may result in the reduced occurrence of blooms in that location.

Table 3. Southern Mosquito Lagoon Emerging Hotspot Analysis classifications.

Pattern	Percent Coverage	Average of Percent Significant Hot Spot	Average of Percent Significant Cold Spot
No Pattern Detected	72%	27.6%	48.4%
Oscillating Cold Spot	4%	12.9%	67.5%
Oscillating Hot Spot	23%	41.9%	33.2%

Table 4. North Indian River Lagoon Emerging Hotspot Analysis classifications.

Pattern	Percent Coverage	Average of Percent Significant Hot Spot	Average of Percent Significant Cold Spot
No Pattern Detected	20%	34.8%	35.7%
Oscillating Cold Spot	75%	25.0%	53.8%
Oscillating Hot Spot	5%	54.4%	18.5%

Within the NIRL there are also several clusters of OHS and NPD, with the majority of the Central NIRL classified as OCS (75%) (Table 4). The OHS are again likely to be locations of shallow SAV which have high estimated ChlA during the summer and then reduced during the winter. However, with an average of 54.4% hot spots and 18.5% cold spots over the time series, this may suggest that the possible SAV in these locations is less impacted by winter temperatures. These clusters are also located next to the natural areas of Merritt Island National Wildlife Refuge and the Kennedy Space Center. The OCS areas were on average 25.0% hot spots and 53.8% cold spots, with NPD at 34.8% hot spots and 35.7% cold spots. The OCS classification of the Central NIRL may be the identification of that portion of the basin having the most bloom activity over the timeseries. The clusters of NPD on the west and east shores of the NIRL may also be indicative of SAV or localized areas of elevated bloom activity over the time series, as these areas cluster around both developed and natural areas.

Within the BR, there are several clusters of OHS and NPD throughout the basin, with 70% of the area being classified OCS (Table 5). 79.3% of significantly increasing patterns were found to be in the North BR. The OHS had an average of 47.3% hot spots and 37.2% cold spots, with one cluster in the North BR along the Canaveral Space Force Base and then another just south of the City of Cocoa Beach. These are also likely locations of shallow SAVs. The OCS clusters have an average of 40.7% hot spots and 48.1% cold spots, while NPD have 39.2% hot spots and 45.0% cold spots. The BR OCS clusters have almost twice the occurrences of significant hot spots when compared to the other basins and are concentrated in the North BR, which aligns with the previous observations that the North BR had the most bloom activity. The four clusters of NPD in the South BR and one in the North BR may be locations of localized bloom activity.

The CIRL is the only basin to have ICS and no OHS or NPD, additionally it is the only area to have significantly decreasing ChlA concentrations from 2015-2021. The majority of the CIRL (82%) (Table 6) was classified as OCS, having 8.4% hot

Table 5. Banana River Lagoon Emerging Hotspot Analysis classifications.

Pattern	Percent Coverage	Average of Percent Significant Hot Spot	Average of Percent Significant Cold Spot
No Pattern Detected	18%	39.2%	45.0%
Oscillating Cold Spot	70%	40.7%	48.1%
Oscillating Hot Spot	13%	47.3%	37.2%

Table 6. Central Indian River Lagoon Emerging Hotspot Analysis classifications.

Pattern	Percent Coverage	Average of Percent Significant Hot Spot	Average of Percent Significant Cold Spot
Intensifying Cold Spot	18%	5.7%	91.2%
Oscillating Cold Spot	82%	8.4%	84.2%

spots and 84.2% cold spots. The ICS areas had an average of 5.7% hot spots and 91.2% cold spots. The least amount of bloom activity was observed in the CIRL in both the literature and SJRWMD monitoring data, corroborating the classifications herein. However, the four distinct clusters of OCS from the City of Melbourne to the north may warrant additional investigation, as they have average percent hot spots of between 12-15%.

4.0 Conclusions and Further Research

4.1 Conclusions. 4.1.2 Sentinel Estimated Chlorophyll A—Within the IRL, the red edge ChlA reflectance bands of the S2 and S3 sensors can accurately estimate most of the variation in ChlA concentrations across the lagoon between 2015 and 2021. The S2 NDCI algorithm had an R^2 of 0.81 and RMSE of 14.14 μg/L for the calibration dataset. The S3 NDCI algorithm had an R^2 of 0.92 and RMSE of 10.01 μg/L for the calibration dataset. The RMSE values calculated for the S2 and S3 NDCI algorithms can be interpreted as the algorithm's ability to create an estimate of ChlA concentrations within 14.14 μg/L for S2 and 10.01 μg/L for S3. Additionally, the high spatial resolution of the S2 imagery highlighted the value of remote sensing of ChlA as concentrations in the surface can be highly variable and surface water grabs may not best represent a given basin.

4.1.3 Harmful Algal Bloom Spatiotemporal Clustering—To assess if HAB intensity, scale, and duration has been increasing throughout the IRL, a Time Series Clustering and Emerging Hot Spot analysis was performed on the S2 estimated ChlA concentrations in the IRL from 2015-2021. The Time Series Clustering analysis identified the North-North IRL and Central-North IRL to have statistically significant increasing trends in ChlA. The remaining basins had increasing but non-significant trends in ChlA.

The Emerging Hot Spot analysis further identified areas of localized HAB activity. This analysis identified the majority of IRL having increasing concentrations of ChlA, however the occurrence of high ChlA concentrations (>40 μg/L) do not appear to be increasing in intensity, scale, or duration as assessed by the Sentinel 2 satellites between 2015-2021. The lack of Intensifying Hot Spots supported this conclusion. Only the CIRL had locations of ICS identified, indicating that the location likely has an increasing frequency of low ChlA concentrations from 2015-2021.

The locations with OHS are likely to be shallow water with SAVs with strong red edge reflectance which reduces during the winter. OCS are locations where ChlA concentrations are usually low but increase in a non-random but likely seasonal pattern with most of these locations occurring in the Central-North IRL and North BR. These may be locations of persistent, seasonal HAB activity. Lastly the NPD locations have a random distribution of hot and cold spots over the time period and as the majority of South BR is classified as NPD it may be described as a basin with sporadic HAB activity.

4.2 Further Research. 4.2.1 Sentinel Estimated Chlorophyll A—The algorithms assessed in this study were calibrated against the available ChlA concentrations measured by the SJRWMD. The ability of these algorithms to differentiate between the various species of HAB-causing phytoplankton in the IRL as identified by Lopez et al., (2021) and Phlips et al., (2021) can be evaluated using both the historical data collected by Florida Fish and Wildlife Commission, Fish and Wildlife Research Institute and future HAB event sampling. The addition of hyperspectral data can be used to further identify peaks that may be more efficient in unmixing the spectral contributions from the water column and benthic environment. Additionally, the potential interference of SAV that was identified in the Emerging Hot Spot analysis concentrations can be further evaluated to remove or mitigate their influence on the observed reflectances.

4.2.2 Harmful Algal Bloom Spatiotemporal Clustering—The clusters and hot spots of historically high ChlA concentrations identified in the HAB spatiotemporal analysis should be used to identify potential causes and further investigate the local patterns of historical ChlA activity. The grouping of Mosquito Lagoon with the North-North IRL, for instance, should be further reviewed to evaluate whether specific patterns of HAB moving from one location to the other, or if they occur simultaneously. Smaller, basin-specific boundary areas for the Time Series Clustering and Emerging Hot Spot analysis should be used to further characterize the division of the BR into north and south segments with several areas of OHS present. With the high performance of the NDCI algorithm for both S2 and S3 the viability of combining these two sensors should be further explored, as well.

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