

# High-resolution shipboard measurements of phytoplankton – a way forward for enhancing the utility of satellite SST and Chlorophyll for mapping microscale features and frontal zones in coastal waters

Christy. A. Jenkins<sup>a</sup>, Joaquim I. Goes<sup>\*b</sup>, Kali McKee<sup>b</sup>, Helga do R. Gomes<sup>b</sup>, Robert Arnone<sup>c</sup>,  
Menghua Wang<sup>d</sup>, Michael Ondrusek<sup>d</sup>, P.V. Nagamani<sup>e</sup>, T. Preethi Latha<sup>e</sup>, K. H. Rao<sup>e</sup>,  
V. K. Dadhwal<sup>e</sup>

<sup>a</sup>Columbia Univ. in the City of New York, New York, NY, 10025, USA;

<sup>b</sup>Lamont Doherty Earth Observatory at Columbia Univ., Palisades, NY, 10964, USA;

<sup>c</sup>Dept. of Marine Science, Univ. Southern Mississippi, Hattiesburg, MS, 39406, USA;

<sup>d</sup>NOAA Center for Satellite Applications and Research (STAR), College Park, MD 20740, USA;

<sup>e</sup>National Remote Sensing Centre, Indian Space Research Organization, Hyderabad, Telengana, 500042, India

## ABSTRACT

Coastal eddies, frontal zones and microscale oceanographic features are now easily observable from satellite measurements of SST and Chl *a*. Enhancing the utility of these space-borne measurements for biological productivity, biogeochemical cycling and fisheries investigations will require novel bio-optical methods capable of providing information on the community structure, biomass and photo-physiology of phytoplankton associated on spatial scales that match these features. This study showcases high-resolution *in-situ* measurements of sea water hydrography (SeaBird CTD<sup>®</sup>), CDOM (WetLabs ALF<sup>®</sup>), phytoplankton functional types (PFTs, FlowCAM<sup>®</sup>), biomass (bbe Moldaenke AlgaeOnlineAnalyzer<sup>®</sup> and WetLabs ALF<sup>®</sup>) and phytoplankton photosynthetic competency (mini-FIRE) across microscale features encountered during a recent (Nov. 2014) cruise in support of NOAA's VIIRS ocean color satellite calibration and validation activities. When mapped against binned daily, Level 2 satellite images of Chl *a*, K<sub>d</sub>490 and SST over the cruise period, these high-resolution *in-situ* data showed great correspondence with the satellite data, but more importantly allowed for identification of PFTs and water types associated with microscale features. Large assemblages of phytoplankton communities comprising of diatoms and diatom-diazotroph associations (DDAs), were found in mesohaline frontal zones. Despite their high biomass, these populations were characterized by low photosynthetic competency, indicative of a bloom at the end of its active growth possibly due to nitrogen depletion in the water. Other prominent PFTs such as *Trichodesmium* spp., *Synechococcus* spp. and cryptophytes, were also associated with specific water masses offering the promise and potential that ocean remote sensing reflectance bands when examined in the context of water types also measurable from space, could greatly enhance the utility of satellite measurements for biological oceanographic, carbon cycling and fisheries studies.

**Keywords:** Satellite, microscale, phytoplankton

## 1. INTRODUCTION

Until a few decades ago, oceanographers relied solely on data collected aboard research cruises, many of which were of limited duration and over small spatial domains. Although data poor, these expeditionary approaches resulted in major advances in our understanding of the global oceans, their productivity and their coupling to physical oceanographic processes and to atmospheric events.<sup>1,2</sup> While these studies expanded our view of oceanographic processes, they also demonstrated the need for sustained sampling over a range of temporal and spatial scales essential for capturing more complex processes necessary to unravel how the oceans operate physically, chemically, and biologically as an integrated system.

The advent of satellite remote sensing of the oceans in the 1970s, and the availability of synoptic maps of ocean features, provided a new means to collect synoptic observations of the global ocean, revolutionizing the way how oceanographers

viewed and studied the oceans.<sup>1,3</sup> With rapid advancements in technology, including the development of novel sensors, improvements in data processing, communication and information transfer methods, etc., oceanographers are now in a position to obtain near real-time, global, high spatial and temporal resolution data of key oceanic variables such as sea surface height and ocean circulation, sea surface temperature, sea surface salinity, ocean color, sea surface winds etc., allowing them to monitor the oceans almost continuously from space.<sup>3</sup>

Within the field of ecosystem dynamics in particular, ocean color satellites have made it possible to closely examine between biogeochemical and physical oceanographic processes such as winter convective mixing, upwelling, mesoscale eddy activity, etc.<sup>4,5</sup> More recent improvements in satellite measurements, in particular improvements in the resolution of satellite oceanographic products now provide biological oceanographers with the capacity to study the distribution of phytoplankton biomass and productivity in relation to small scale microscale physical oceanographic features.<sup>6,7</sup>

While it is known that sub-mesoscale physical processes such as micro-scale eddies, shingles, frontal zones etc. can create patchiness in phytoplankton biomass, they are also responsible for creating ecological niches, that are many a times dominated by a single or a few phytoplankton groups that that need to be appropriately represented and accounted for before the full potential of satellite ocean color data for carbon cycling studies is realized.<sup>6,8,9</sup> Information of phytoplankton functional types (PFTs) and /or phytoplankton size classes (PSCs) is not only essential for capturing the complexity of carbon cycling and carbon export in the oceans, but are a pre-requisite for constraining ecosystem models.<sup>10</sup>

While there has been progress in estimating PFTs and PSCs from space, lack of data to validate ocean color derived PFT or PSC products continues to remain a major impediment towards further progress.<sup>9,11-14</sup> Most ocean color PFT and PSC validation efforts are either based on algorithm inter-comparisons or on ecosystem based expectations of phytoplankton community structure; for instance, the expectation that diatoms dominate the community during upwelling events<sup>9,14,15</sup> which are not necessarily accurate at all times.

Here we showcase results from a recent cruise to the mid-Atlantic Bight that demonstrate the utility, the potential and promise of PFT and PSC measurements using continuous sampling methods. During our cruise we integrated several state-of-the-art optical instruments capable of measuring PFTs and PSCs into the ship's continuous flowing uncontaminated seawater system. Subsets of the data acquired by each of the instruments are presented. This study is not aimed at providing a comprehensive analysis or a detailed account of phytoplankton community structure or discusses the data from an ecosystem dynamics viewpoint, but is rather an attempt to illustrate the richness of the datasets collected by the approach proposed for validating satellite PFT and PSC measurements, especially in physically dynamic and ecologically complex coastal water masses.

## 2. MATERIALS AND METHODS

Our study was carried out on board the NOAA Ship *Nancy Foster* during a 14 day cruise (9-22 November, 2014) in the western Atlantic along the USA mid-Atlantic Bight. The primary purpose of this cruise was to collect *in-situ* optical and ancillary data for validation of JPSS VIIRS satellite ocean color radiometry and derived products.<sup>16-18</sup> In addition to sampling at fixed stations, the ship's seawater flow-through system was equipped with several bio-optical and hydrographic instruments for continuous underway sampling. The intake point for seawater into the sea chest was at a depth of 3m. All of the instruments listed below were connected in parallel and were synced in time and location with the ship's Global Positioning System while continuously sampling the coastal waters of the mid-Atlantic Bight. The ship track was adjusted and optimized continuously to transect microscale physical oceanographic features of the Gulf Stream front and offshore that were discerned from daily images of SST and Chlorophyll *a*.

The instrument suite included the following:

1. Two ac-9s (WET<sup>®</sup> Labs), one with a filter and the other without, for determining total absorption, CDOM absorption, total scattering, beam attenuation, and particle absorption at 9 wavelengths,<sup>19</sup>
2. ECO BB3 (WET<sup>®</sup> Labs) for backscattering at 3 wavelengths (469, 529 and 652 nm),
3. Conductivity and temperature sensor for estimating SST and seawater salinity,
4. Fluorometer for chlorophyll measurements,<sup>19</sup>
5. FlowCAM (Fluid Imaging Technologies<sup>®</sup>, Inc.) for dynamic imaging particle analysis for species composition and size measurements,<sup>20</sup>

6. Advanced Laser Fluorometer - ALF (WET<sup>®</sup> Labs) for determinations of CDOM and for characterizing phytoplankton functional groups based on chlorophyll and phycobilipigments,<sup>21,22</sup>
7. Mini Fluorescence Induction and Relaxation (FIRE) for phytoplankton photosynthetic competency, non-photochemical quenching,<sup>23</sup>
8. AlgaeOnlineAnalyzer (bbe Moldaenke<sup>®</sup>) for determination of Chl *a*, and major phytoplankton groups.<sup>24</sup>

Along the cruise track 23 stations were occupied for collection of water samples from discrete depths and for underwater profile measurements of ocean color and biology. For the purpose of the present study, we focus on the data collected by instruments listed from 3 to 8 that provided measurements of phytoplankton species composition and biomass, phytoplankton size, and photosynthetic competency. Data from instruments 1 and 2 and from the discrete stations are not part of this study. Instruments 3 and 4 are used routinely on board research vessels for measurements of SST, salinity and fluorometric Chlorophyll *a*. Measurements made by instruments 5 to 8 are described in detail below:

### 2.1 Automated Laser Fluorescence (ALF) measurements of phytoplankton groups

The ALF combines high-resolution spectral measurements of blue (405 nm) and green (532 nm) laser-stimulated fluorescence with spectral deconvolution techniques to quantify fluorescence of Chl *a* (peak at 679 nm), three phycobilipigment types: PE-1 (peak at 565 nm), PE-2 (peak 578 nm) and PE-3 (peak at 590 nm), Chromophoric Dissolved Organic Matter (CDOM, peak at 508 nm) and variable fluorescence (Fv/Fm). All fluorescence values obtained are normalized to water Raman spectra and generally expressed as relative fluorescence units (RFU), whereas Fv/Fm is unitless. PE-1 type pigments are associated with blue water or oligotrophic cyanobacteria with high phycourobilin/phycoerythrobilin (PUB/PEB) ratios, PE-2 type phytoplankton with low-PUB/PEB ratios are generally associated with green water cyanobacteria that usually thrive in coastal mesohaline waters, and PE-3 attributable to eukaryotic photoautotrophic cryptophytes. RFU values for Chl *a* can be converted into mg m<sup>-3</sup> Chl *a* values using least square regressions of acetone or HPLC measured Chl *a* with RFU values for Chl *a* measured in an ALF.

### 2.2 FlowCAM measurements phytoplankton identification, cell counts and cell sizes

The FlowCAM is a particle imaging system that can be used for imaging and counting of phytoplankton in seawater. The instrument provides information on the total number of particles imaged per volume of sample, together with the dimensions of each particle allowing estimations of phytoplankton community structure and particle size distribution of both phytoplankton and of detrital particles. On board the NOAA ship *Nancy Foster*, the instrument was equipped with a 4X objective (UPlan FLN, Olympus<sup>®</sup>) and a 300 µm field-of-view flow cell which allowed taxonomic identification and sizing of phytoplankton greater than 10 µm in size. The instrument was programmed to sample seawater over 20 min intervals with a break of 1 min to allow cleaning of the flow cell in between runs. Assessments of particle shapes, size and names of phytoplankton up to the genus level were undertaken back in the shore laboratory with the Visual Spreadsheet program (v. 2.2.2, Fluid Imaging).

### 2.3 FIRE measurements of photosynthetic competency

The FIRE technique was developed to measure a comprehensive suite of photosynthetic and physiological characteristics of photosynthetic organisms<sup>23,25</sup>. This technique provides a set of parameters that characterize photosynthetic light-harvesting processes, photochemistry in Photosystem II (PSII), and the photosynthetic electron transport down to carbon fixation. Because these processes are particularly sensitive to environmental factors, the FIRE technique can be utilized to provide a measure of natural (nutrient limitation, photoacclimation and photoinhibition, thermal and light stress, etc.) and anthropogenic stressors (such as pollution). One property that is unique and the most sensitive to environmental stressors is Fv/Fm. All optical measurements by the FIRE are sensitive, fast, non-destructive, and can be done in real time and *in-situ* and can provide an instant measure of the photosynthetic competency of the cells

### 2.4 The AlgaeOnlineAnalyser (AOA)

The AOA allows for continuous measurements of fluorescence. The sample is excited with light from colored light emitting diodes (370nm, 470nm, 525nm, 570nm, 590nm and 610nm) and the resulting excitation spectra are utilized for estimating Chlorophyll *a* and taxonomic discrimination of PFTs, i.e. cyanobacteria, green algae, brown algae (diatoms and dinoflagellates) and cryptophytes on the basis of the shapes of the excitation spectra.

### 3. RESULTS

The cruise track covered a distance of 1800 km, and was optimized to transect and sample transient microscale features such as shingles, filaments and meanders in the offshore region, and cross-shelf features such as upwelling and downwelling zones, coastal fronts, etc. in the coastal zone (Fig 1). Four major water masses were encountered along the cruise track as seen in the T-S plot (Fig. 2). CDOM values measured by the ALF have been incorporated into this plot to highlight the origins (riverine, coastal, upwelled and open ocean), and optical characteristics of different water masses of the four water types. The coldest and least saline waters were encountered off Cape Hatteras which comes under the influence of Pamlico Sound estuarine complex. These waters were also extremely CDOM rich indicating a rich terrestrial source for CDOM. Mesohaline, low temperature, CDOM rich waters were also encountered off Charleston Harbor region and at several locations close to the coast of South Carolina. Upwelled waters were distinguishable from the coastal waters on the basis of their high salinity (35 to 36) and low temperatures (18 to 20°C). These were also CDOM rich.

In Figs. 3a-c, we have used datasets from the ALF to illustrate the differences in the community composition within the four major masses delineated above. Fig. 3a shows that the PE-1 type communities of open-water *Synechococcus* spp. and *Trichodesmium* spp. were more closely associated with the high temperature and high salinity waters of the Gulf Stream.<sup>21</sup> This is not unexpected as both *Synechococcus* spp. and *Trichodesmium* spp. thrive in waters such as the Gulf Stream that are usually poor in nitrogenous nutrients. On the other hand, the low salinity, low temperature riverine waters were dominated by PE-2 type communities comprising of the coastal water *Synechococcus* spp. PE-3 type phytoplankton which are usually dominated by cryptophytes were predominant in the mesohaline coastal waters. These organisms usually thrive in low salinity waters that have low, but not limiting concentrations of nutrients.

Variable fluorescence values (Fv/Fm) obtained continuously over the entire period of the cruise using the mini-FIRE provide a measure of the photosynthetic competency of the dominant phytoplankton communities (Fig. 4a).<sup>23</sup> With the exception of the communities of diatoms and Diatom-Diazotroph Association (DDA) which dominated the upwelling zone, Fv/Fm values revealed communities of phytoplankton that were photosynthetically active and healthy throughout our study area. The low Fv/Fm (<0.3) of the diatoms (Fig. 4b) and DDAs (data not shown) are indicative of population whose further growth was being limited by nutrient availability.

Although four major water types were encountered during the cruise, the steep, and at times abrupt changes in seawater salinity (from 32.4 to 37.4 psu) and temperature (14.5 to 27.1°C) visible in the distance plots along the cruise track are indicative of the presence of microscale features, fronts and upwelling zones, suggestive of the highly dynamic and complex nature of the physical oceanographic conditions in this region (Fig. 5a).

Chlorophyll *a* monitored along the entire cruise track (Fig. 1) revealed that these microscale features, fronts and upwelling zones were sites of active phytoplankton growth (Fig. 5b). Chlorophyll *a* concentrations (as indicated by arrows in Fig. 5b) were always higher within these features. The highest Chlorophyll *a* concentrations were recorded in the upwelling zone between 33.5 and 34°N Lat., south of Cape Fear, North Carolina. Other locations where Chlorophyll *a* concentrations were high were the coastal upwelling region off Charleston, and the frontal zone off Cape Hatteras.

Continuous underway monitoring by the bbe AOA, the FlowCAM and the ALF aided in teasing out different PFTs associated with these regions of elevated Chlorophyll *a* concentrations. Chlorophyll concentrations associated with different PFTs, show that Diatoms were the dominant phytoplankton group in regions of upwelling, whereas green algae were the only PFT present in the upwelled water off Charleston. Cryptophytes on the other hand, were present throughout the coastal region, but were especially high within the microscale features (Fig. 6a-c).

The spatial distribution patterns of PFTs shown in Fig. 7, which is based on the data from the FlowCAM, is indicative of large differences in phytoplankton communities along the coast. For instance DDAs, diatoms and cryptophytes were clearly the dominant PFTs in the upwelling region south of Cape Fear. On the other hand, cryptophytes made up a significant fraction of the phytoplankton population in the upwelling zone off Charleston. On the other hand, diatom concentrations were high in the frontal zone off Cape Hatteras. In the purely low-Chlorophyll *a* Gulf Stream waters, *Synechococcus* was the dominant phytoplankton. In other regions within the Gulf Stream that revealed the presence of streamers and filaments, diatoms appeared to be the dominant PFT.

## 4. DISCUSSION

It is becoming increasingly apparent that accurate determinations of PFTs and PSCs are essential to developing a mechanistic understanding of global ocean productivity and carbon export. Recent studies,<sup>10,26</sup> showed that most carbon export models are remarkably sensitive to PSCs and PFTs and accurate determination of both are essential for further progress in our understanding of ocean carbon cycling and biogeochemical processes. The need for development of robust and reliable satellite based products of PFTs and PSCs that can be incorporated into future carbon cycling modeling scenarios is clearly an area of emphasis in NASA's recent community supported EXPORTS plan.<sup>10</sup>

One of the biggest challenges facing ocean biogeochemists is that most of the common approaches for measuring PFTs or PSC are either difficult to undertake at sea or time consuming, such that they cannot provide data at the requisite scales necessary to capture the complexity of oceanic ecosystems. Coastal ecosystems in particular are notoriously more complex and are also highly dynamic. Large changes in PFTs and PSC associated with fluctuations in physical and chemical oceanographic conditions that take place over the course of the day are difficult to capture by microscopy and other conventional shipboard techniques.

For this reason, biological oceanographers are increasingly turning towards satellite ocean color data to surmount this perennial problem of PFTs and PSCs under-sampling. In the past few years, there have been several attempts to obtain information on PFTs and PSCs using satellite ocean color data. Despite the progress, routine application of these methods and algorithms for discriminating PFTs and PSC from space have been hampered by the lack of a requisite number of spectral wavebands essential for development of robust algorithms and furthermore by the absence of shipboard *in-situ* datasets of PFTs and PSCs. Most methods that use ocean color data rely on either model to model inter-comparisons or are based on ecosystem traits that contribute to the dominance of one PFT over the other (such as diatom dominance in upwelling regions). In a few instances comparisons have been based on HPLC pigment data,<sup>12,14</sup> which have problems of their own.

The launch of hyperspectral sensors on missions like PACE and multispectral sensor planned for GEOCAPE<sup>27</sup> are expected to significantly improve efforts to discriminate PFTs and PSCs from space. In coastal waters in particular these upcoming missions are expected to allow monitoring of phytoplankton biomass at finer spatial scales than ever before. What will be required is a rich database of shipboard data, to ensure that satellite derived products are accurate and robust over large space and time scales. Underway measurements also offer an opportunity to investigate sub-pixel variability in PFTs and PSCs, whose implications for ocean color measurements from space have not been a topic of discussion, for want of adequate field datasets. Since there is no sacrifice of data quality, underway measurements also offer the opportunity for several fold more matchups, than those possible using conventional shipboard sampling approaches.

The continuous underway sampling methods that we have proposed here clearly are a way forward for mapping PFT and PSC changes associated with microscale features and frontal zones and enhancing the utility of satellite ocean color datasets for ecosystem and biogeochemical studies in complex coastal waters.

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## Figures and captions

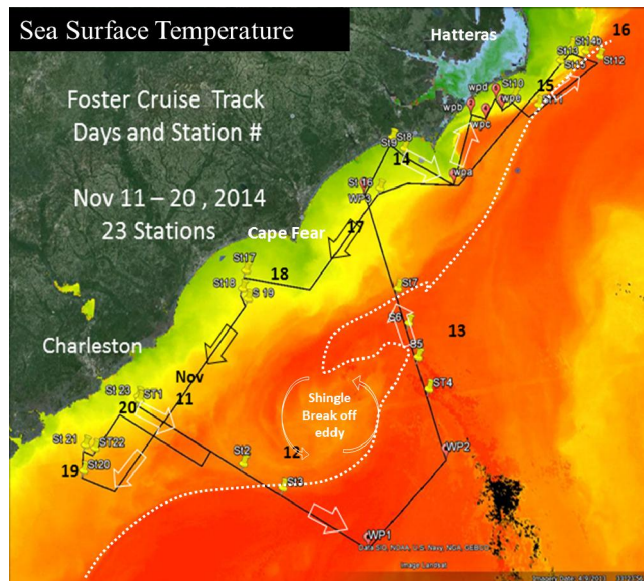


Figure 1. Cruise track of R/V *Nancy Foster* and locations of stations occupied overlaid on image of SST during the period from November 11 to 20, 2014. Arrows indicate onward (white) and return (black) journey of cruise. The white dotted line demarcates the front between coastal and Gulf Stream waters. Also visible in the SST image is the location of a shingle

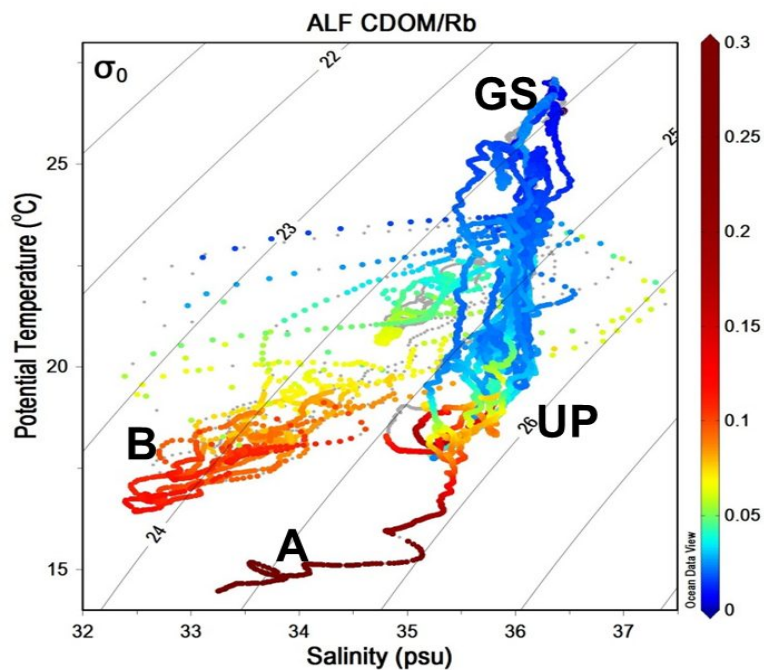


Figure 2. Temperature versus salinity plot of the underway data showing the four major water masses, i.e., GS- Gulf Stream waters, UP – upwelled waters, A – river water influenced waters off Cape Hatteras and B – coastal waters. CDOM concentrations associated with these water masses are shown in color.



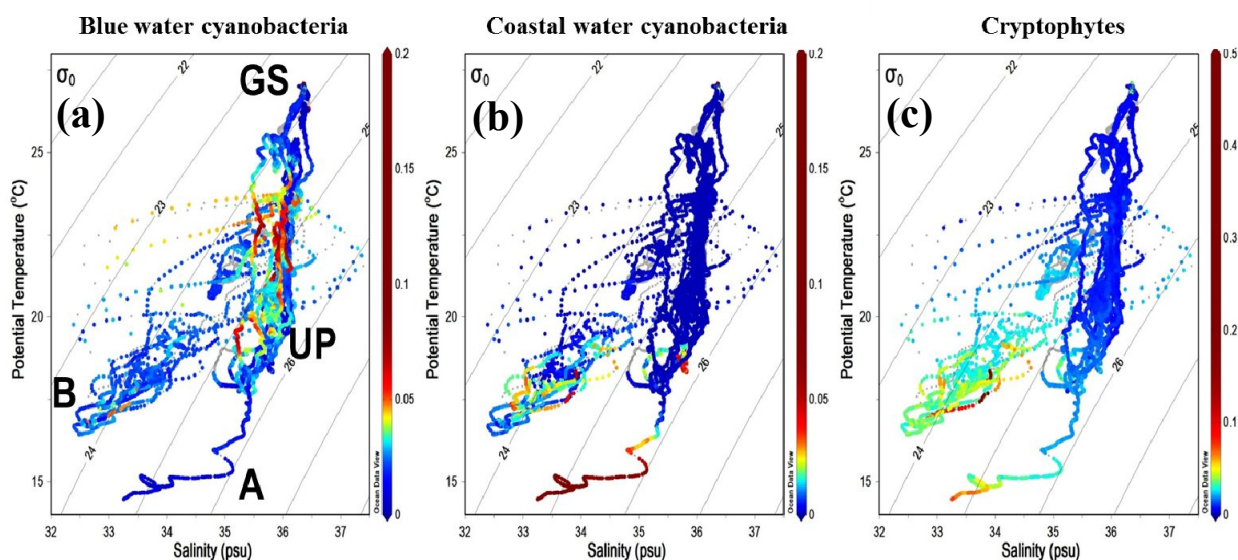


Figure 3. Temperature vs salinity plots showing the distribution of PFTs, a) blue water cyanobacteria, b) coastal water cyanobacteria and c) Cryptophytes derived using the ALF.

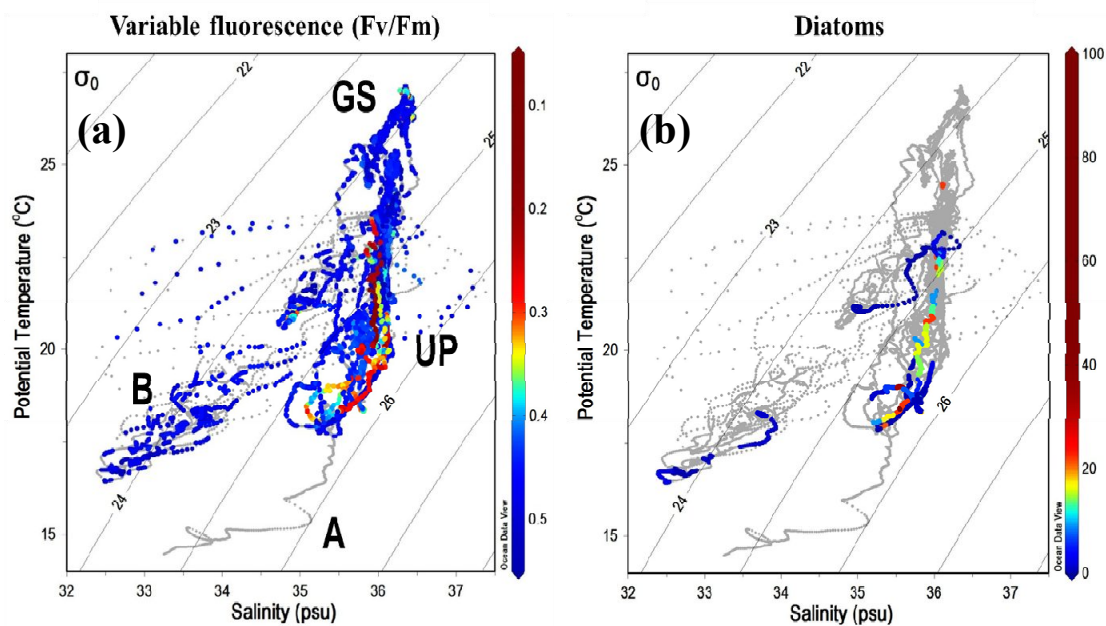


Figure 4. Temperature vs salinity relationships depicting a) photosynthetic competency and b) distribution of diatoms in different water masses encountered during the cruise.



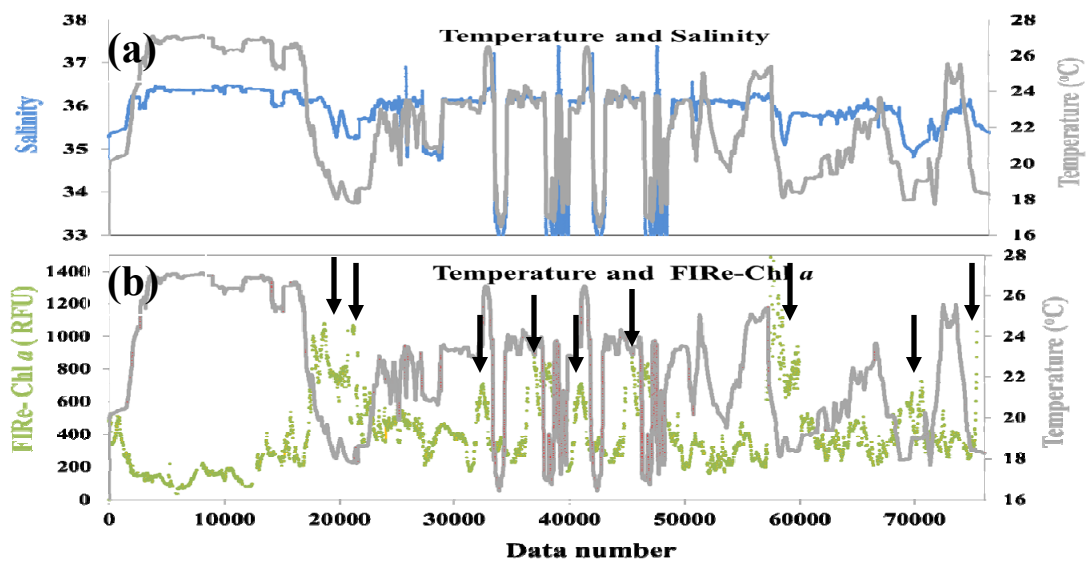


Figure 5. Distance plots showing variations in surface seawater a) temperature and salinity and b) temperature and Chlorophyll *a* along the cruise track. Arrows indicated locations of elevated Chlorophyll *a* along the cruise track.

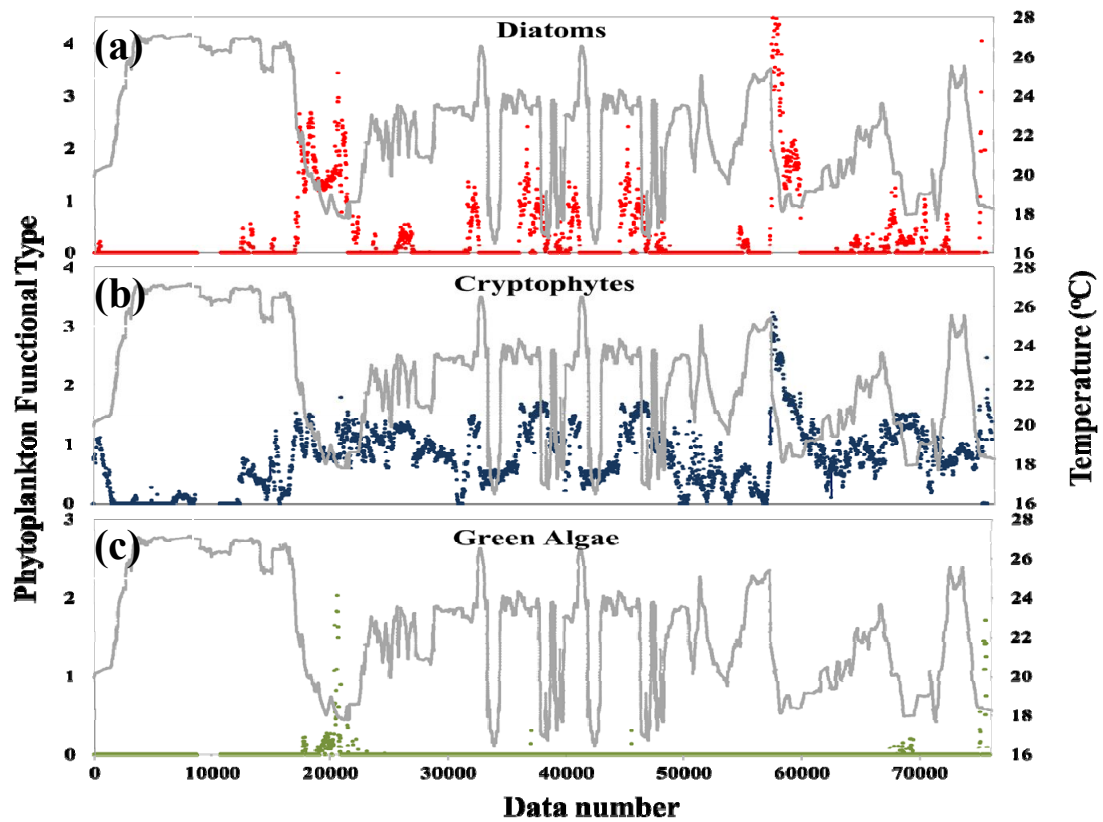


Figure 6. Distance plots showing variations of a) Diatoms b) Cryptophytes and c) Green algae with respect to temperature along the cruise track

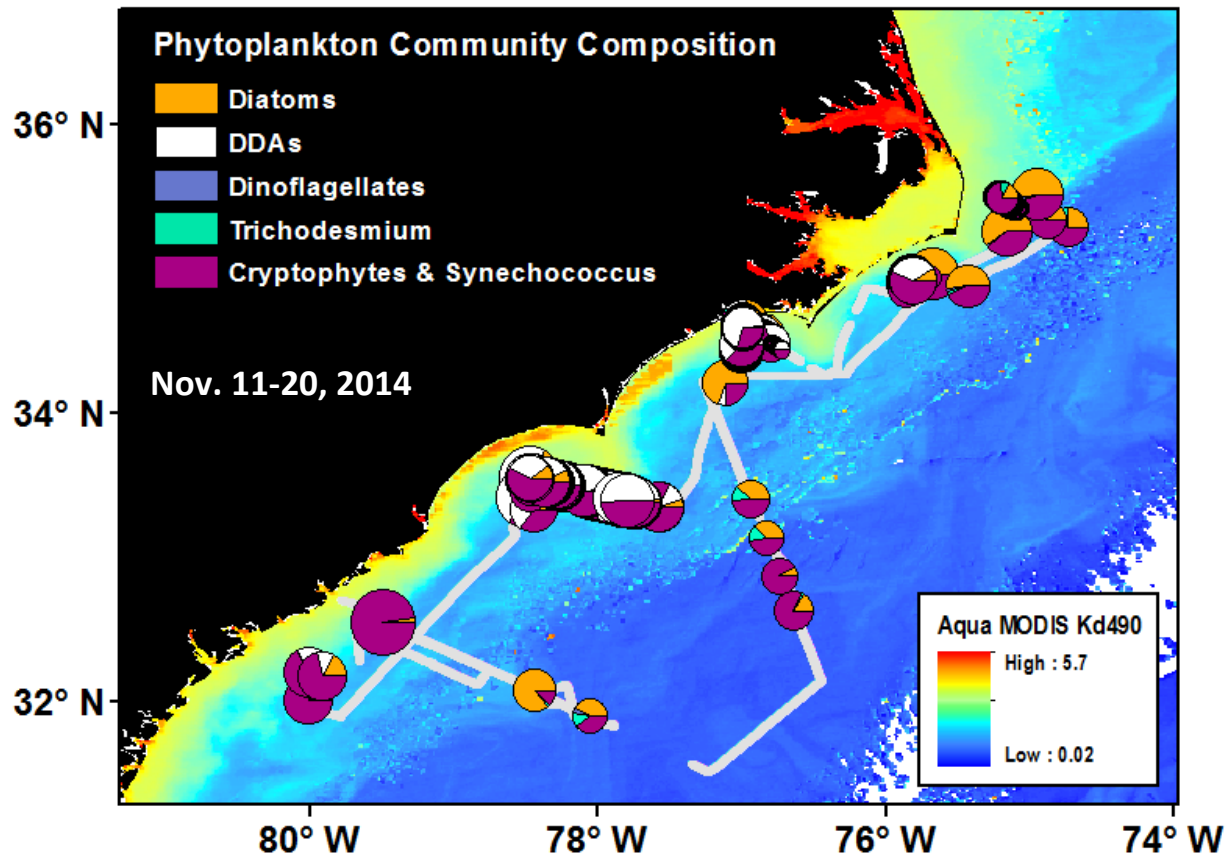


Figure 7. Distribution of PFTs along the cruise track, superimposed on Chlorophyll *a* composite derived by binning daily images of Chlorophyll *a* from VIIRS for the cruise period. Regions of elevated Chlorophyll *a* associated with coastal upwelling in the region south of Cape Fear, in the frontal region near Cape Hatteras and microscale features such as shingles, filaments etc. seen along the boundary of the Chlorophyll *a* rich coastal waters are also visible. PFT data plotted in this figure were derived from the FlowCAM

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