

CHAPTER ONE

INTRODUCTION

1.1 Background

There are a lot of biotic and abiotic components in this earth that create dependency to each other. Our world comprises about 71% of water of which 97.5% salt water. Phytoplankton is called the foundation of aquatic food web as well as the major contributor of global carbon absorption and sequestration to the deeper layer of the vast ocean. About half of the total photosynthesis in the world are performed by marine phytoplankton (Baumerta *et al.*, 2008). Photosynthesis of phytoplankton change inorganic carbon to a fundamental piece of Earth's carbon cycle. Marine ecosystems significant atmospheric CO₂ sink and consume equal levels of CO₂ as terrestrial ecosystems, actually responsible for the elimination from the atmosphere of almost one third of anthropogenic CO₂ emissions (Mackey, 2018). The activities of phytoplankton mainly occur within first 50 meters from the surface but vary from place to place due to turbidity and other nutrients availability (Moore and Abbott, 2000). Similar to terrestrial plants Phytoplankton plays a single key role in ocean carbon cycle and photosynthesis (Strzeppek *et al.*, 2019) through photosynthesis Phytoplankton uptakes atmospheric carbon-di-oxide and turns it into their food and produce oxygen. This carbon-di-oxide is then sequestered to the deep of the water column just like a biological pump and react with the water and form carbonic acid and the sinking carbon by plankton particles accumulate in the bottom. A highly diverse variety of phytoplankton is a microscopic photosynthesizing group of microalgae and cyanobacteria that act as a link between atmospheric and oceanic processes. By fixing about 50 Gt (gigga tonnes) of carbon per annum, Phytoplankton contribute almost 50% to Earth's total primary production (Mackey, 2018).

A set of processes known as the biological carbon pump transports sinking carbon particles from the surface to the deep ocean in a process known as marine snowfall when plankton dies or is consumed. Sinking rates of phytoplankton assemblages with diverse and variable taxonomic composition, growing under a variety of environmental conditions, due to its impact on both the vertical distribution of phytoplankton biomass and the carbon budget of the photographic field, phytoplankton settling has

received substantial attention in the oceanographic literature (Bienfang, 1982). Although plankton cannot regulate its movements against the current, by increasing its surface area-to-volume ratio and producing and releasing lipids, they are able to control their buoyancy (fats) (Greenwell *et al.*, 2010). Colony formation in phytoplankton, according to Stokes' law, lead to increased sinking rates and greater losses of sedimentation if colonies have the same densities as the phytoplankton cells they contain (Peperzak, 2003). There are many classes of marine phytoplankton and their sinking velocity as well as capability are quite different from each other. The importance of understanding the dynamics of sinking phytoplankton is centered on its role in the vertical flow of organic matter in the sea. Phytoplankton sinking may affect the vertical distribution of the plant Biomass and productivity at sea (Szyper, 2014). Success of the phytoplankton population depends on its ability to strike a favorable balance between cell division rates, grazing losses and sinking. For having specific weight of phytoplankton it tends to sink from the euphotic zone to the depth as phytoplankton itself can't move distinctively. By the way it depends on various factor like motility, cellular morphology and many water quality parameters (Greenwell *et al.*, 2010). Diatom sinking rates in rapidly growing cells, however, are independent of cell size over a range greater than $106 \mu\text{m}^3$ (Brad, 2016).

Chlorophyll a is a measure of the abundance of phytoplankton and biomass in coastal and estuarine waters (Desortová, 1981). Phytoplankton sinking rate is mainly depends on chlorophyll-a (Bienfang, 1981). There are many typical chlorophyll concentration determination techniques: spectrophotometry, fluorometry, and high-performance liquid chromatography (HPLC) but a homogenous technique of chlorophyll-a measurement is used widely named SETCOL (Settling Column) and to date, phytoplankton sinking studies have been limited to unialgal populations (Bienfang, 1979). SETCOL method is a simple and reliable method for measuring phytoplankton's sinking rate is defined. Because this technique is suitable for both heterogeneous field populations and unialgal in vitro cultures. The method includes the use of settling columns originally containing a uniformly mixed cell population and calculates a decline rate based on the shift in cell vertical distribution after a given time (Bienfang, 1981). On the other hand, the percentage of chlorophyll content in phytoplankton biomass has been found to vary considerably with the season. The

contents of chlorophyll per unit biomass in the same location may vary by more than two orders (Desortová, 1981).

“Biological pump” is the most widely used biological term in relation to Phytoplankton carbon flux from the surface to the depth. Sinking rate of phytoplankton in water column is associated with phytoplankton carbon flux. Oceanic carbon cycle is a major part of the global carbon cycle and through carbon flux about 30-50% of global carbon di oxide is being sequestrated to the depth of the ocean annually. Plankton transports gases and nutrients from the surface of the ocean into the deep, rather than like a pump (Basu and Mackey, 2018). In the carbon cycle, phytoplankton is quite different from that of trees and other land plants, which actually absorb CO₂ and serve as a carbon storehouse or sink. Instead, during photosynthesis, ocean living particles absorbs CO₂ and, while most gas escapes within about a year, some of it is transported to the depth through dead plants, body parts, feces, and other sinking materials. The CO₂ is then released into the water as the materials decay, and most of it is absorbed by chemically combining with water molecules in the sea water (H₂O). Although a small but possibly significant percentage of the sinking organic material is buried in the ocean sediment, through ocean currents, most of the dissolved carbon dioxide is eventually returned to the surface but this can take centuries or millennia. The composition of the sinking particles may have an impact on both the magnitude and efficiency of the surface carbon flux ocean through the mesopelagic zone and deep ocean (Durkin, 2016). Research shows that average particulate Organic Carbon concentration in euphotic zone is higher during summer and monthly POC (Particulate Organic Carbon) and chlorophyll-a is not correlated

Several research works were performed (Lee *et al.*, 2020, Guo *et al.*, 2016) on carbon sequestration of global carbon cycle and world ecosystem. A new research always brings something new for the further work. Phytoplankton is the key component of the vast oceanic ecosystem and absorb 30-50% of carbon annually which is almost equivalent to the terrestrial plants. In this recent years climate change is being a thread not only for Bangladesh but also for the earth. Increasing amount of carbon di oxide traps global temperature and hamper ecosystem (Fang *et al.*, 2018). Due to having a vast sea water body attached to our main land the water body plays a very important role in ecosystem. But in the eastern Bay of Bengal still there is no work on

phytoplankton sinking rate. As a result there is no secondary data of phytoplankton sinking rate as well as carbon flux in Bangladesh. So, it is going to be a pioneer research in this field. Chlorophyll-a, sinking rate and carbon flux have been estimated in the three selected stations situated in the north-east Bay of Bengal.

1.2 Statement of the problem:

Bangladesh possess about 711 km long coastal line and a vast water body along the Bay of Bengal. Day by day the word “blue economy” is being spread worldwide. The Blue Economy conceptualizes oceans and seas as “Development Spaces” where spatial planning integrates conservation, sustainable use of living resources, oil and mineral wealth extraction, bio-prospecting, sustainable energy production and marine transport. Blue Economy offers a suite of opportunities for sustainable, clean, equitable blue growth in both traditional and emerging sectors like Fisheries, Shipping and Port Facilities, Aquaculture, Tourism, Biotechnology and marine genetic resources etc. But Bangladesh is still more lagging behind in this field due to lack of preliminary research and survey on Bay of Bengal. In general there are some studies on phytoplankton or zooplankton abundance, seasonal variation to nutrients availability (Ahmad, 2019). There is no report on phytoplankton sinking rate and seasonal variation of carbon absorption. As phytoplankton contribute almost 50% to Earth's total primary production (Mackey, 2018) it is needed to gather seasonal variation of carbon absorption by phytoplankton of the Bay of Bengal for giving a clear concept of oceanic environment which is also very essential in the field of Marine fisheries.

1.3 Significance of the study:

Though phytoplankton is the major weapon in oceanic ecosystem to flux atmospheric carbon to the deep of the ocean to make a buffer condition in the atmosphere so it is essential to know the features of carbon flux. This research has been conducted in the coastal area of some oceans even most of the coastal part of Bay of Bengal. But this research still not conducted in north-east part of Bay of Bengal. As environmental condition and geographical location of north-east part of Bay of Bengal is different from other oceans where this research has been conducted so the findings of this research may differ from the other. In respect of Bangladesh this kind of data are not still exist to the researcher. I will study the composition of phytoplankton and identify

which major phytoplankton species are the main contributor for daily and seasonal carbon sequestration

1.4 Objectives of the research work:

- To perceive seasonal variation of chlorophyll-a and phytoplankton sinking rate of major groups of phytoplankton
- To understand variation of carbon flux associate with phytoplankton sinking rate in three stations of North-east coast of Bangladesh
- To justify the correlation between carbon sinking rate and nutrients availability

CHAPTER TWO

REVIEW OF LITERATURE

Increasing carbon is a threat for the globe in this recent years. A lots of relevant studies are being conducted around the world to get a concept about how much carbon are emitted and how much are being absorbed by plant sources either higher class or lower (Bond *et al.*, 2013). According to Goeppert *et al.* (2012) earth is going to a vulnerable condition and atmospheric carbon is the major reason of anxiousness. So, it cannot be denied the contribution of photosynthesis by plant or oceanic phytoplankton in absorption of this huge carbon from the atmosphere. According to Baumerta (2008) about half of the total photosynthesis in the world are performed by marine phytoplankton and it is about 50 Gt per annum through carbon fixation which is the research outcome of Mackey (2018).

As sinking of phytoplankton is the vehicle of atmospheric carbon to the deep water, in many countries this research has been conducted. In very earlier period of this field Eppley *et al.*, (1967) tried to estimate marine phytoplankton sinking rate with a Flucometer method. They found that non growing culture cells sink four times faster than growing culture cells. He also found that this organism's sinking rate was much greater than that predicted from its cell.

After 10 years according to Bienfang *et al.* (1977) sinking rate of marine phytoplankton in a technologically modified and improved method named discrete sample layer (DSL) method. They mainly used Homogenous Sample (HS) method and so called t₀-5 technique to estimate the mean sinking rate to know if any cell in the population sink twice faster than the expected average sinking rate. They were satisfied with their applying method and believed that these theoretical and technological advances make it possible to predict mean sinking rates more easily and with greater precision and accuracy than previous approaches.

In 1979, Bienfang et al introduced a very effective method which was more accurate than the previous one. He described the method which was based on detection of radioactively (¹⁴C) labeled phytoplankton acquired from a productivity-type incubation. The system overcame a variety of technological and theoretical problems previously prohibited by precise sinking normal phytoplankton populations' rate assessments. The distinctive characteristic of this approach was that the biomass

parameter was monitored ^{14}C is indexed. This referred to a greater sensitivity than previous approaches and also included a parameter that described a sample's photo synthetically active elements.

Till now the most acceptable method of phytoplankton sinking rate was introduced by named the SETCOL method. It was found that this method was very suitable for both heterogeneous field populations and unialgal *in vitro* cultures. The approach included the use of settling columns originally comprising a uniformly mixed cell population and calculated a sinking rate based on the shift in cell vertical distribution of cells after a given time. From changes in the vertical distribution of biomass, this system produced both sinking rate and ascent rate values (Bienfang, 1981). In this research this SETCOL method was used.

The most importance of this study is Carbon flux and sequestration through the phytoplankton sinking. Here the study can connect between sinking and carbon flux in an oceanic environment. Around the world a lots of research are found related to carbon sinking through phytoplankton from the surface to the deep of the ocean. Parvez *et al.* (2018) conducted a research on the factor effecting phytoplankton carbon flux in marine environment. His analyzed data indicated that the variable production ratios of biogenic silica to POC in different ocean regions were responsible for the poor correlation observed between silica and POC in deep sediment traps, and that the homogeneous ratios of calcium carbonate to POC observed in these same traps could be caused by high concentrations of suspended coccoliths in deep waters.

There was conducted a precise research on Carbon flux through Bacterioplankton. To test the microbial loop hypothesis by observing the fate of carbon-14 labeled Bacterioplankton for over 50 days, an enclosed water column of 300 cubic meters was used by them. They found that only 2 per cent of the label initially carbon-14-labelled glucose was found in larger species after 13 days, at which time about 20 per cent of the overall label added remained in the particulate fraction. Factors effecting carbon flux also studied by Laura *et al.* (2008) in NW Mediterranean coastal site through bacterial activity. Bacterial growth efficiencies were measured by short-term and long-term methods and ranged from 3 to 42%, increasing during phytoplankton blooms in the winter (during Chl-a peak) and in the spring. Changes in the structure of the bacterioplankton assemblage (as depicted by denaturing gradient gel

electrophoresis fingerprinting) had not been coupled with changes in the functioning of the ecosystem, at least in bacterial carbon use.

Due to parameters variation carbon sinking rate also vary. According to Guo *et al.* (2016) phytoplankton sinking rate associated with carbon sinking rate in the Changjiang (Yangtze River) estuary was approximately 2.4 times higher that in summer during *P. dentatum* bloom (average = $26.10 \pm 26.25 \text{ mg C m}^{-2} \text{ days}^{-1}$) in spring. Except for temperature and nitrite concentration in summer, no significant correlation was found between phytoplankton sinking rates and most of the environmental parameters in this research. Field measurements showed that differences in sinking could be correlated with changes in concentrations of irradiance and nitrate. Since these variables had no direct effect on water density, physiological changes affecting cell buoyancy had to be induced. Although a direct response to a single environmental variable was not always apparent, the rate of decline was positively correlated with the rate of growth in the marginal ice zone, implying a connection to physiological processes. Pitcher *et al.* (1989) studied on the topic of southern Benguela upwelling system Phytoplankton sinking rate dynamics also using the SETCOL method. It was found that sinking rates were affected by the length of the Settling checks. The sinking rate of chlorophyll ranged from 0 to 0.91 m per day but was poorly associated to phytoplankton carbon sinking rates which ranged from 0 to 0.78 m per day. Sinking rates of phytoplankton populations were not significantly correlated to any of the assessed environmental parameters, but were significantly correlated with taxonomic features of the assemblages that were regulated by the prevailing climate.

Brad *et al.* (2016) found that during rapid sinking, nutrient flow in large diatoms increased and current mass transport models did not incorporate the unstable sinking behavior observed in this research. This study showed that swimming or sinking, however, could dramatically increase a cell's nutrient uptake rate.

The sinking rates of phytoplankton were determined by different environmental parameters such as temperature, irradiance and level of nutrients duing the study of Bienfang (1984). Studies on the connection between the sinking of phytoplankton ratings and concentration of nutrients showed that the former is closely related to link to the latter, with a nutrient limitation that often results in increased rates of sinking (Kilham, 1976). According to Guo *et al.* (2016) contrary to expectations, during these

two dominant organisms (*Prorocentrum dentatum* and *Skeletonema dorhnii*) there was no association between cell size and sinking rate. The found value of three nutrients NO₂-N, PO₄-P and SiO₃-Si were 0.359, 0.108 and 0.673 respectively during spring and 0.535, 0.608 and 0.096 during summer

There is no study related to phytoplankton sinking rate and carbon sinking in north-east Bay of Bengal so primary data are being missed. But lots of research conducted mainly on Phytoplankton community, nutrients dependency and all other parameters associate with the phytoplankton composition along the coastal line of Bay of Bengal.

Sarwar *et al.* (2010) conducted a research on the Karnafully River Chittagong, Bangladesh which included water parameters. The mean total suspended solids concentration was found 365 mg/L in the Karnafully river estuary ranged from 120 to 590 mg/L. The mean total concentration of dissolved solids was found 8018 mg/L, ranging from 292 to 18530 mg/L. The pH value was in Karnafully river estuary was ranged from 6.2 to 7.0 and the mean salinity was found 4.8 mg/L with a range from 0.4 to 9.2 mg/L. Another research done by Ahmad (2019) in Karnafully river estuary included major plankton groups and nutrient gradients from season to season and station to station. During this research about 13 species of phytoplankton were found among these 5 major species were *Coscinodiscus sp*, *Chaetoceros sp*, *Nitzschia sp*, *Pseudonitzschia sp* and *Pleorosigma sp*. Phytoplankton sinking has a relation to phytoplankton community structure and phytoplankton community depends on nutrients availability (Guo *et al.*, 2016)

Parvez *et al.* (2018) conducted a study on phytoplankton assembles and their hydrological factors in the Rezu khal estuary which was adjacent to main Bay of Bengal water body. A total of 27 genera of Phytoplakton fewer than 4 divisions were identified during the study period. During the current study, certain dominant genera were found to be *Biddulphia*, followed by *Coscinodiscus*, *Rhizosolenia*, and *Nostoc*.

After going through all the related papers of home and aboard it is clear that though a lots of study on phytoplankton sinking rate and carbon fluxing have been conducted in foreign countries but still such kind of research are not conducted in Bangladesh. During this research key emphasis given on phytoplankton sinking rate, carbon absorption, Chlorophyll-a concentration, major species responsible for carbon sinking and relationship between sinking and nutrients availability in three specific stations

(Teknaf, Patenga and Bashbaria). It has covered both the Teknaf and Chattagram coast. The north-east part of Bay of Bengal is different from the north-west part of Bay of Bengal on the basis of environmental factors. As a result phytoplankton sinking rate also would be different from the existing result found of these area. In this research two major seasons were selected because of lacking of enough funding. Moreover in Bangladesh winter and monsoon are the mostly dominant seasons. However this research hopefully will play as a key research in the field of phytoplankton carbon sinking for the further research.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Research extends:

Bay of Bengal is the Northern arm of Indian Ocean, part of Bay of Bengal Large Marine Ecosystem (BoBLME) which is connected with 8 countries of both south and south East Asia. Bangladesh is also one of them which is situated on the north east coast of Bay of Bengal. The research was carried out in three definite stations alongside the Teknaf to Chattogram coast in monsoon and winter. Those specific stations were selected on the basis of water parameters, anthropogenic exposure and tidal conditions.

Here is the name of those 3 stations with their geographical locations

Station 1: (S1). Bashbaria coast ($22^{\circ}36'40.09''\text{N}$ $91^{\circ}37'32.05''\text{E}$)

Station 2: (S2). Potenga ($22^{\circ}13'3''\text{N}$ and $91^{\circ}47'11''\text{E}$)

Station 3: (S3). Teknaf coast ($20^{\circ}50'48''\text{N}$ and $92^{\circ}16'24''\text{E}$).

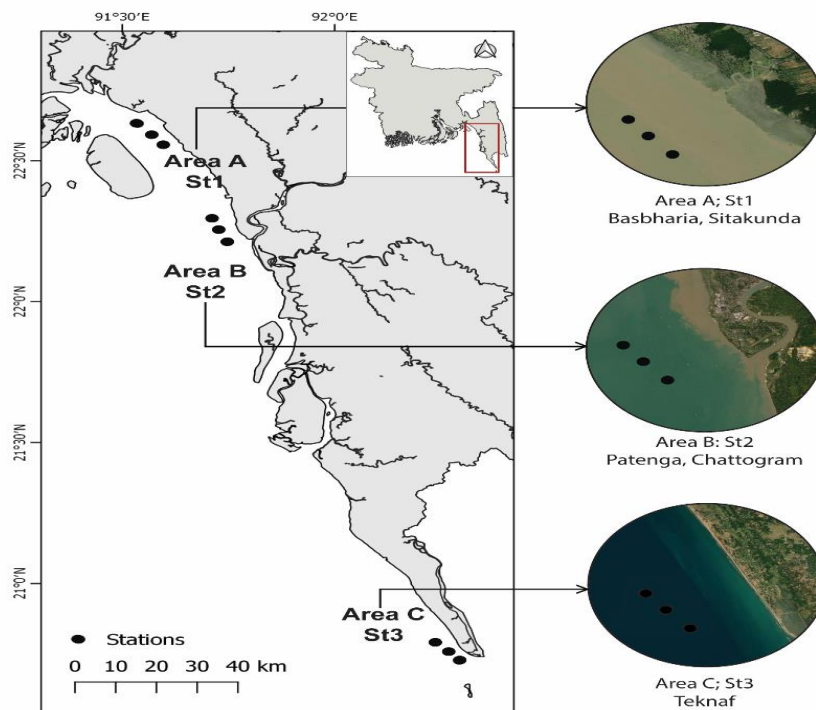


Fig. 1: sampling locations in three stations (Bashbaria,patenga and Teknaf)

3.2 Sampling:

3.2.1 Water collection for SETCOL and parameter testing:

This research was based on the depth-wise sinking rate of phytoplankton and chl-a measurement of this specific depth was must for calculating sinking rate. So, water had to collect from 3 depths of the water column so sample was collected by a Nansen water sampler bottle using a hired boat. Water collection was done at high tide due to have phytoplankton abundance. Sample was collected from surface, 5 meter and 10 meter depth from 3 points of a specific station keeping at least 10 meter distance from one to another for escaping similarity. The physical parameters that pH, salinity and temperature were measured immediately on boat. After that samples were kept in black bottle for escaping photosynthesis and filtered within 2 hours. Sample was collected in two seasons (monsoon and winter) from 3 respected stations



Fig. 2: Sample collection by Nansen water sampler

3.2.2 Water collection for phytoplankton composition and abundance:

Water was further collected using phytoplankton net for phytoplankton composition and observed under microscope in oceanography laboratory under the department of Marin Bio resource Science of Chattogram Veterinary and Animal Sciences University. At first the net was dipped down in the water up to 10 meter depth. Then filtered residue was collected in a bottle immediately that sample was preserved by giving 3-4 drops 10% formalin that was previously made and brought to the laboratory. Three replications were done for increasing accuracy and sample was collected in two seasons (monsoon and winter) from 3 respected stations.



Fig. 3: Phytoplankton sample collection by plankton net

3.3 Analysis of bio chemical parameters:

Seasonal distinction of physical and bio chemical parameters of 3 stations were carefully measured by following standard procedures and using supporting devices

3.3.1 Analysis of physico-chemical water quality parameters:

Water quality parameters like temperature (Celsius Thermometer), pH (Portable pH meter), and salinity (Refractometer) were monitored from each station in two seasons. Three replication of water samples were collected.

3.3.1.1 Total Suspended Solid (TSS):

Total suspended solids were determined by the filtration procedure (Rhodes, 1985) following a filtration protocol. Water samples were filtered through glass fiber filters that were dried at 105 ° C (> 1 hr) and weighted to obtain the sum of suspended solids for turbidity determination (TSS).

Equipment's: Filter paper, electrical balance, oven, Desiccator

Method:

- a. The filter paper was first dried in the oven and put in the Desiccator (at least 30 min at both stage). Oven-dried filter paper was then weighted.
- b. 50 ml of sample water taken and filtered with filter paper.
- c. Filter paper dried in the oven at 104 °C after filtration and put at Desiccator
- d. Then weight of the filter paper with the remaining solids was calculated.

- e. Finally TSS in a sample of water was calculated using the following formula.

Calculation:

$$\text{TSS} = \frac{B-A}{50} \times 1000$$

Where,

A= Weight of the oven dried filter paper

B = Weight of the filter paper with remaining solid.

3.3.1.2 Nitrite-nitrogen (NO₂-N):

Nitrite-nitrogen (NO₂-N) was determined following the methods described by Bend (White *et al.*, 1952)

Equipment's:

Spectrophotometer (Model: Osk-15745), funnel, conical flask, measuring cylinder, filter paper.

Method:

- a. The 50 ml water sample was filtered by the Watman filter paper (0.1µm).
- b. The 50 ml filtered sample was then stored in a conical flask.
- c. 1 ml of added and mixed sulphanilamide.
- d. Provided for 2-8 min.
- e. Then 1ml of NNED was added.
- f. Estimated the extinction at 543 nm after 10 minutes but prior to 2hrs.
- g. (µg at NO₂-N/Kg): Factor (19.84) X (Absorbance of samples – absorbance of blank).

3.3.1.3 Phosphate-Phosphorus (PO₄-P):

Phosphate-Phosphorus (PO₄-P) was determined following the methods described by Murphy *et al.* (1961)

Equipment:

Spectrophotometer (Model: Osk-15745), funnel, conical flask, measuring cylinder, filter paper.

Method:

- 1) A 50 ml OF water sample was filtered by the Watman filter paper (0.1 μ m).
- 2) Then 50 ml of the filtered sample was taken in a conical flask.
- 3) Added 2 ml of ammonium molybdate were added and shaken.
- 4) Then 5 drops of stannous chloride was added.

5) Finally, the absorbance of the developed color measured at 690 nm in a Spectrophotometer (Model: Osk-15745)

Calculation:

(μ g at PO₄-P/Kg): Factor (45.93) x (Absorbance of sample – absorbance of blank)

3.3.1.4 Silicate-Silicon (SiO₃-Si):

Silicate-Silicon (SiO₃-Si) was determined following the methods described by Mullin and Riley (1955).

Equipment's:

Spectrophotometer (Model: Osk-15745), funnel, conical flask, measuring cylinder, filter paper.

Reagents:

10% Acid ammonium molybdate, 25% Sulphuric Acid.

Method:

- a. A 50 ml water of sample was filtered by the Wasman filter paper (0.1 μ m).
- b. Then 50 ml of the filtered water sample was taken in a conical container.
- c. 2 ml of ammonium molybdate added and shaken.
- d. 0.5 ml of Sulfuric Acid was added.
- e. Finally, the absorbance of the developed color measured at 460 nm.

Calculation:

(μ g at SiO₃-Si /Kg)= Factor (5372.58) x (Absorbance of sample – abs. of blank).

3.4 SETCOL:

SETCOL is a reliable and technologically simple method for measuring phytoplankton sinking rates. SETCOL stands for Settling Column. It is a cylinder like structure made by PVC and mouth opening covered by a transparent circular plate. The diameter of the mouth is 2.5 cm and the height is 0.6 meter (60 cm). Each cylinder has 3 chambers of different volume like 100 ml(upper),1000 ml (middle) and 100 ml (bottom). For each depth water sample settling three SETCOL bottles were needed and total 9 for 3 depths of a particular station. Three bottles are fixed with a strong structure made either by iron or wood.

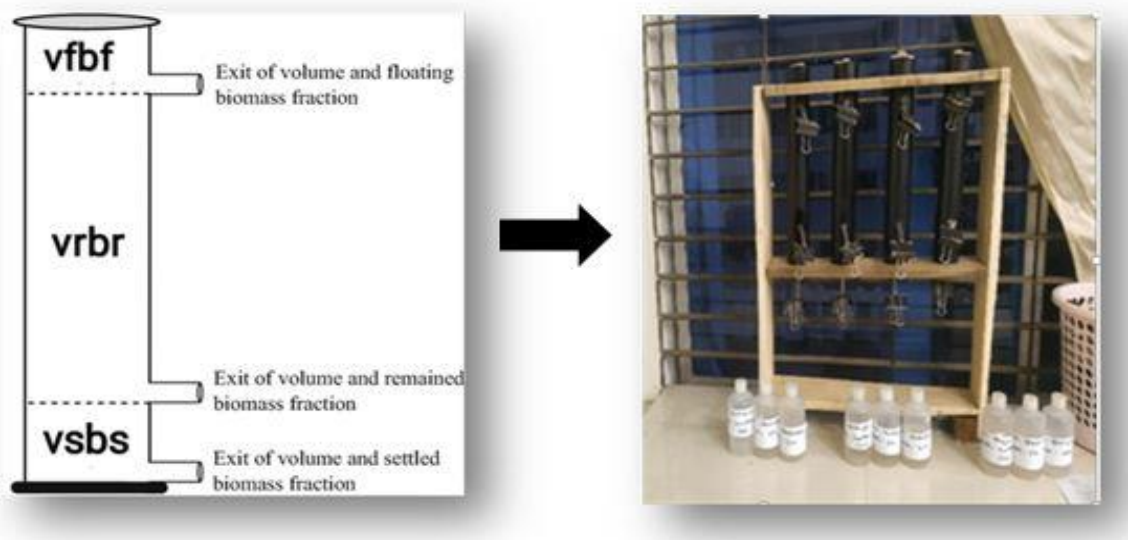


Fig. 4: SETCOL Bottle (1.source: Google)

3.5 Chlorophyll-a measurement:

For most photosynthetic species, Chlorophyll-a is essential for the release of chemical energy. It is also a key component of phytoplankton sinking rate measurement. But in this research Chlorophyll-a was estimated following an established SETCOL method (Bienfang, 1981). A long procedure was followed which was slightly different from the conventional chl-a measurement. At first water samples collected from a station immediately was taken to the laboratory as soon as possible. After that SETCOL bottle were filled with the sample water of 3 depths and kept around 2 hours for settling. For each depth three bottles were used and total nine bottles were used for 3 depth of a particular station. After 2 hours settled water samples were taken from each bottle. Though each bottle had 3 chamber so there were about 9 separate samples for

each depth and total 27 separated samples for 3 depth. After that the following steps were followed:

Using a vacuum pump, water samples were filtered by a membrane filter (0.45 μ m) of 3 volumes (100 ml, 1000 ml and 100 ml). The filtered membranes were moved to 10 ml of 90% acetone and kept overnight. Using a glass rod, the filtered documents were thoroughly mixed with acetone. Then the centrifugation was carried out for 2.30 minutes at 3500 RPM. , Compared to blank acetone, the supernatant contents (extract) were taken into corvettes and the extract absorbance was estimated at 664, 647, 630 and 750 nm in Mecasys Spectrophotometer by Optizen. Final chlorophyll-a concentration was calculated according to following equation (Talling and Driver, 1963). The absorbance at 750 nm was subtracted from those three wavelengths (Johan1 *et al.*, 2015).

$$\text{Chlorophyll-a} = (11.85 A_{(664-750)} - 1.54 A_{(647-750)} - 0.08 A_{(630-750)}) * (V/S) * 1000$$

Where,

- A_{664} = Absorbance at 664 nm
- A_{647} = Absorbance at 647 nm
- A_{630} = Absorbance at 630 nm
- V = Volume of acetone used (ml)
- S = Volume of sampled filter (ml)



Fig. 5: Chlorophyll- a estimation

3.6 Phytoplankton sinking rate measurement:

Sinking rates of particulate matter were determined using a homogeneous sample method called SETCOL (Bienfang 1981). To determine phytoplankton sinking rate the average chl-a of 3 chamber of the SETCOL bottle were calculated first. Then the following formula was used to determine sinking rate per hour and convert to one day by multiplying 24 hours.

$$\text{Sinking rate} = \text{Bs/Bt} * \text{L/t}$$

Here,

Bs= The average biomass of phytoplankton settled in bottom

Bt = The average total biomass of a column

L=Length of this column

T=Time period of settling

3.7 Qualitative and quantitative estimations of plankton

A 45 µm mesh sized phytoplankton net was used for getting concentrated phytoplankton sample. At first the net was put down up to 10 meter under water with binding a weight stone for sinking. Then the net was pulled toward the surface and concentrated sample was taken and preserved with 5% buffered formalin. Three samples were taken in same way from different points for getting a homogenous sample and directly brought to laboratory for both quantitative and qualitative estimation of phytoplankton.

Using a Sedgwick-Rafter cell Cell containing 1000 1mm³ cells qualitative and quantitative estimations of plankton were done. A 1ml sample was taken in the S-R cell and left for 15 minutes undisturbed to allow plankton to settle. The plankton in 10 randomly selected cells were identified up to family level and counted under a binocular microscope with imaging facilities. The planktons were also observed under microscope to study the major plankton classes. In the S-R cell, a 1ml sample was taken and left undisturbed for 15 minutes to allow the plankton to settle. The plankton was classified up to genus level in 10 randomly selected cells and counted with imaging facilitated computer under a binocular microscope. To find major phytoplankton group qualitative estimation of phytoplankton was done by identifying and classifying. There was also three replication for accuracy. Plankton abundance was calculated by using this formula:

$$N = (P * C * 100) / L$$

Where,

- N = Number of Plankton cells or units per liter of original water (Counted by using Sedgwick-Rafter cell)
- P = The number of plankton counted in 10 fields
- C = The volume of final concentration of the sample (ml)
- L = The volume (L) of water sample

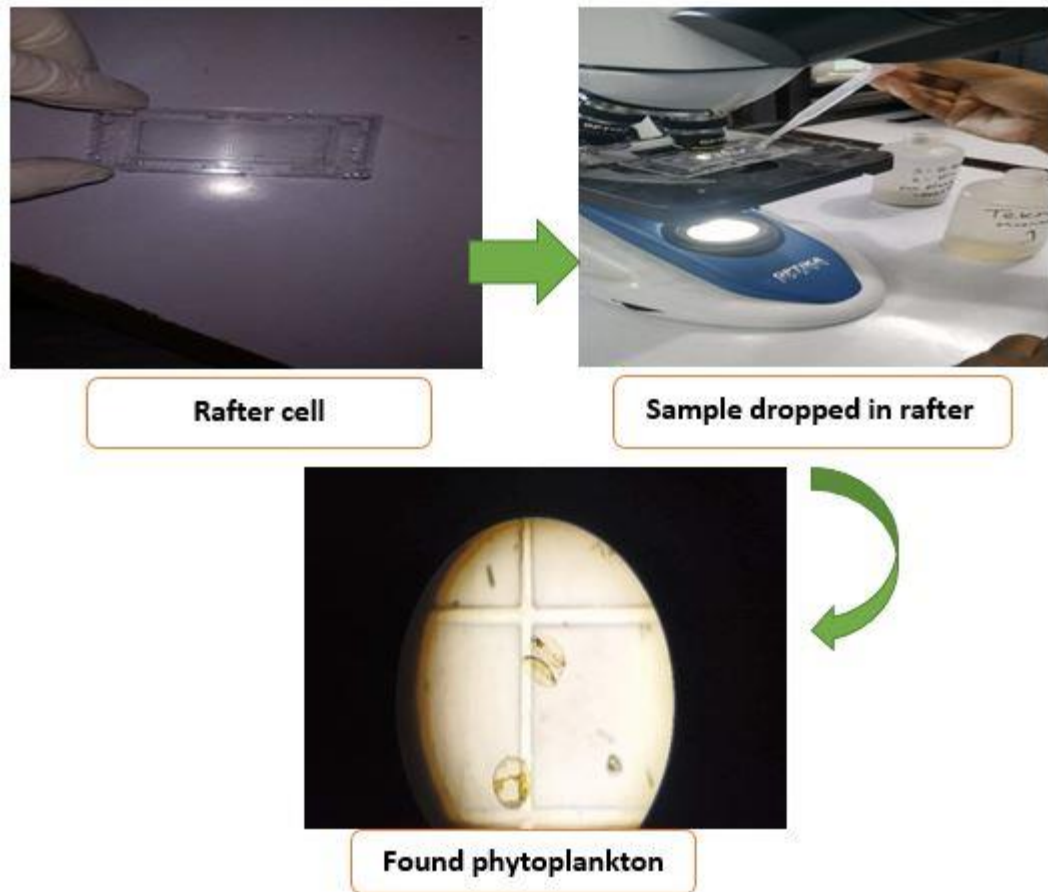


Fig. 6: Phytoplankton counting and identification

3.8 Carbon flux determination:

Carbon flux is the carbon sequestration from one depth to another via sinking particles like phytoplankton in ocean. During this research carbon flux was estimated by the multiplication of sinking rate and total carbon of phytoplankton according to Guo *et al.* (2015)

3.8.1 Total carbon estimation in each cell and in a specific depth:

Cell carbon was estimated according to Guo *et al.* (2015) also. To calculate this firstly phytoplankton cell diameter, length and width were measured by a micrometer scale under the microscope which was facilitate with a computer (Irina, 2006). After that cell volume was calculate geometrically (Irina, 2006). Then the carbon/cell was calculated using volume of each plankton. There were used two formula, one for diatom and another for algae (Guo *et al.*, 2015).

1. $\text{Log}_{10} C = 0.76 \times \text{Log}_{10} V - 0.352$ for diatom

2. $\text{Log}_{10} C = 0.94 \times \text{Log}_{10} V - 0.60$ for algae

Here,

- C is the carbon content of each species in pg C/cell
- V is the cell bio volume of each species in μm^3

Carbon content in each cell further multiplied with total cells found in a depth of one specific phytoplankton. Thus total amount of carbon in a specific depth was calculated and finally carbon flux was calculated by this formula (Guo *et al.*, 2015)

❖ **carbon flux= sinking rate \times total carbon of a specific depth**

According to Beinfang, 1981

$$\text{Sinking rate} = \text{Bs/Bt} * \text{L/t}$$

3.9 Data Analysis:

The water quality data for each station were analyzed on seasonal basis and the findings were demonstrated using Microsoft Excel 2013. Pearson correlation was done with multiple factors using two-way ANOVA with SPSS version 22.0.0 and Principle Component Analysis was done among stations and seasons using R-software.

CHAPTER FOUR

RESULTS

4.1 Physico-chemical parameters:

The water quality parameters of the selected stations were recorded over two seasons. These physical-chemical parameters included temperature, salinity, pH, TDS, Water depth and nutrients included $\text{NO}_2\text{-N}$, $\text{PO}_4\text{-P}$, $\text{SiO}_3\text{-Si}$ and Chlorophyll-a. All the value was replicated 3 times for more accuracy.

4.1.1 Temperature:

Temperature is regarded as a vital parameter of water which is related to most of the parameters and varies from season to season. During this study the highest temperature (32.3°C) was found in monsoon at Patenga station in the depth of 5 meter and the lowest 23.2°C during winter in tekna. The average value found in Bashbaria was 30.5°C (monsoon) and 25.03°C (winter), in Patenga was 32.4°C (monsoon) and 26.4°C (winter), in Teknaf was 32.0°C (monsoon) and 23.6°C (winter). Two-way ANOVA results showed that variations in water temperature among 3 stations and 2 seasons were significant ($p < 0.05$)

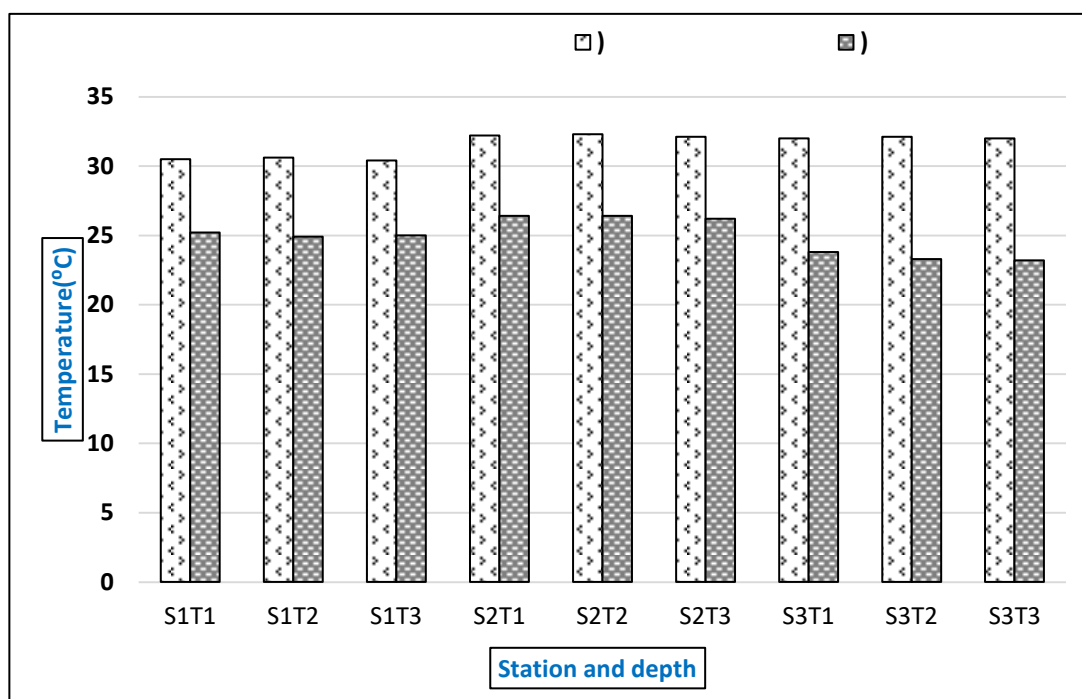


Fig. 7: Average temperature fluctuation among 3 stations

4.1.2 Water pH:

Water pH was comparatively higher in winter and gradually decreased towards monsoon in each station. No significant fluctuation was observed. The highest Water pH 7.6 in Bashbaria during winter in the depth of 10 meter and the lowest was observed 6.3 in Teknaf during monsoon in the depth of 10 meter. The average value found in Bashbaria was 6.9 (monsoon) and 7.5 (winter), in Patenga was 6.7(monsoon) and 7.1(winter), in Teknaf was 6.3 (monsoon) and 6.5 (winter). A Two-way ANOVA showed that variations in water pH among 3 stations and 2 seasons were significant ($p < 0.05$).

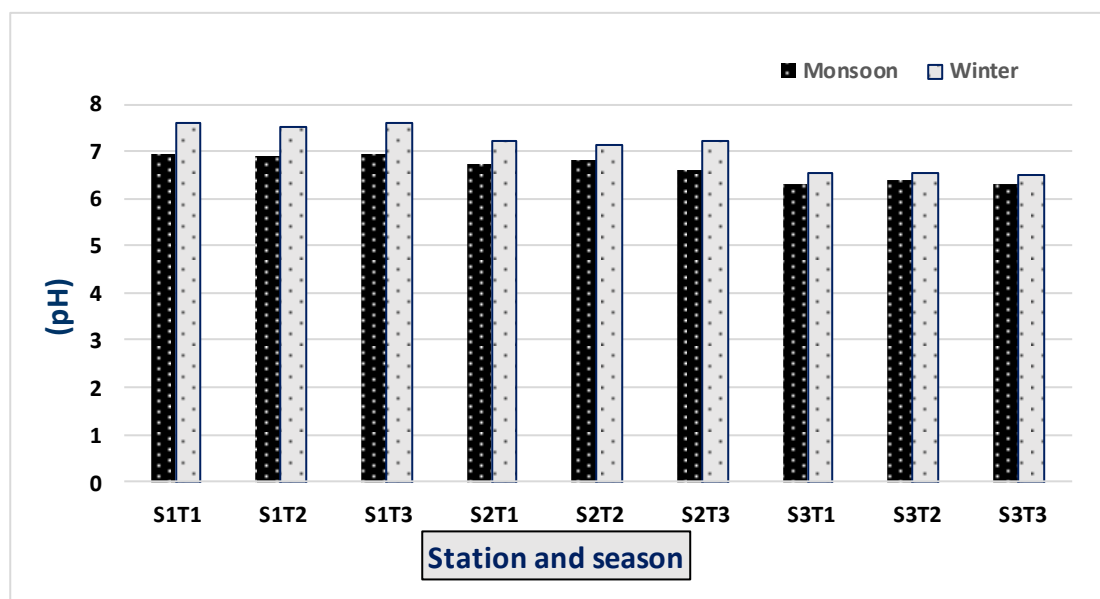


Fig. 8: Average pH fluctuation among 3 stations

4.1.3 Salinity:

During the study salinity was homogenous in the most of time except monsoon at Patenga. It was observed drastically fall of salinity in three depths during monsoon. However the highest salinity was 33 ppt at Teknaf during winter and lowest was 3 ppt in Patenga during monsoon. The average value found in Bashbaria was 10.1 ppt (monsoon) and 16.5 ppt (winter), in Patenga was 3.6 ppt (monsoon) and 21.3 ppt (winter), in Teknaf was 25.7 ppt (monsoon) and 32.7 ppt (winter). A Two-way ANOVA showed that variations in water salinity among 3 stations and 2 seasons were significant ($p < 0.05$)

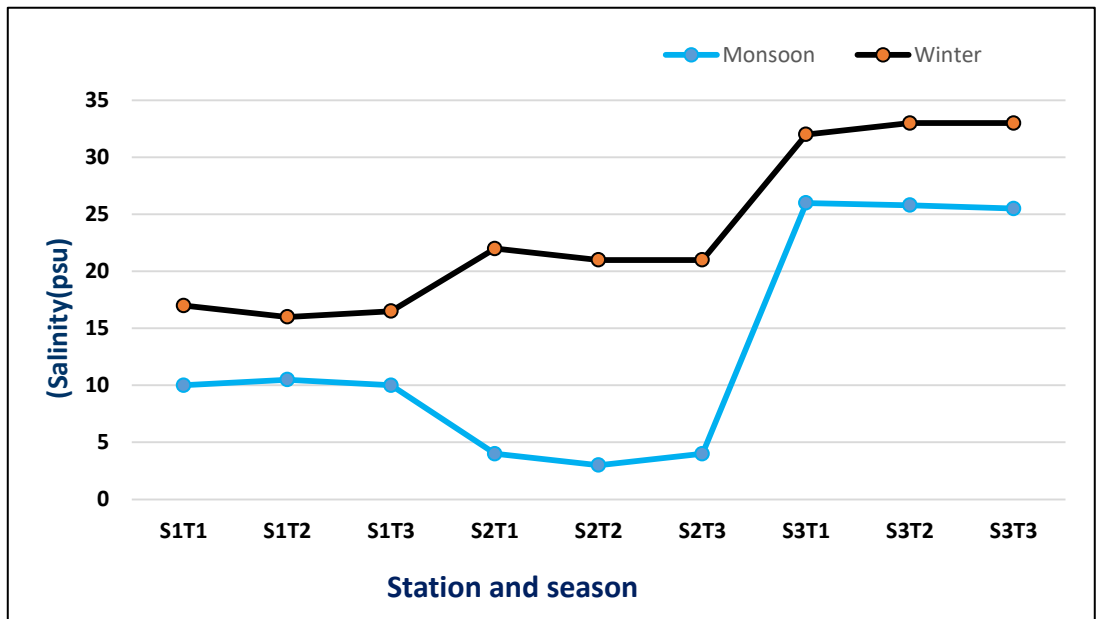


Fig. 9: Average Salinity fluctuation among 3 stations

4.1.4 Total Dissolve Solid (TDS) and Total Suspended Solid (TSS):

TDS was changed as the salinity changed in water column. The highest value of TDS was observed as 30.6 g/L during winter at Teknaf station in the surface layer and the lowest TDS was 2.8 g/L during monsoon at Patenga in 5 meter depth. However the average TDS value found in Bashbaria was 11.13 g/l (monsoon) and 17.06 g/l (winter), in Patenga was 2.82 g/l (monsoon) and 22.3 g/l (winter), in Teknaf was 24.66 g/l (monsoon) and 30.5 g/l (winter). A Two-way ANOVA results showed that variations in TDS among 3 stations and 2 seasons were significant ($p < 0.05$)

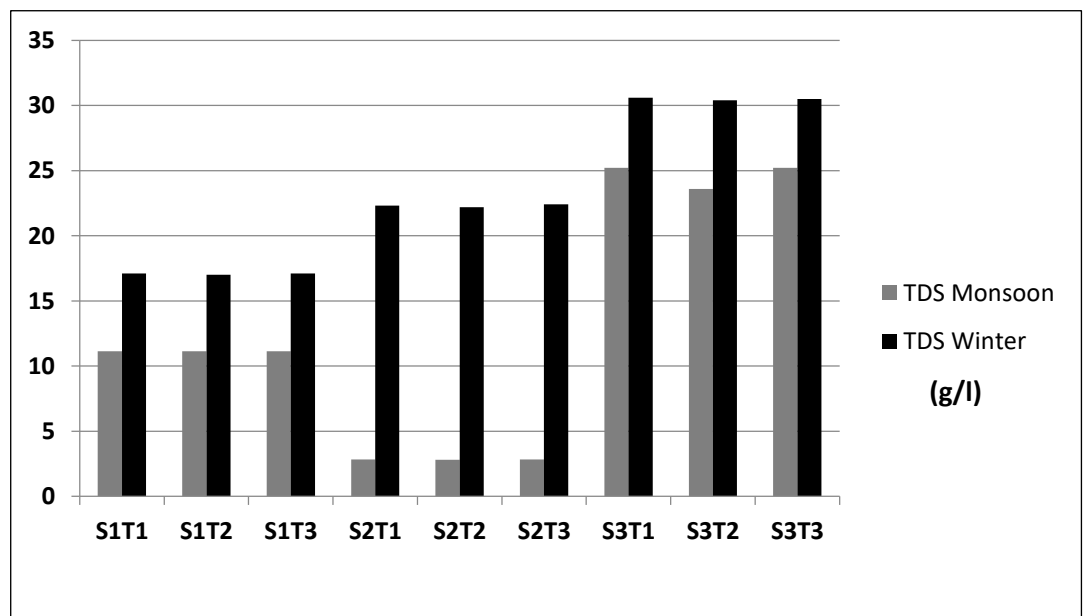


Fig. 10: Average TDS fluctuation among 3 stations

On the other hand the average TSS value found in Bashbaria was 0.30 g/l (monsoon) and 0.84 g/l (winter), in Patenga was 0.29 g/l (monsoon) and 0.94 g/l (winter), in Teknaf was 0.31 g/l (monsoon) and 0.57 g/l (winter). Two-way ANOVA results showed that variations in TDS among 3 stations and 2 seasons were significant ($p < 0.05$)

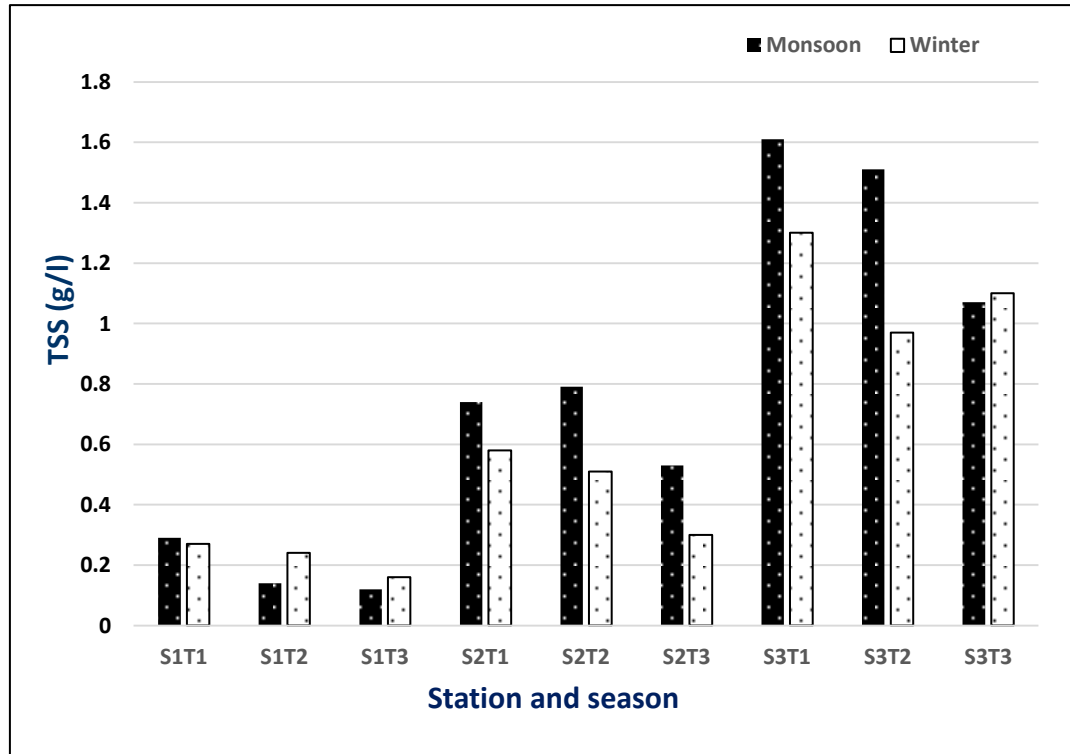


Fig. 11: Average TSS fluctuation among 3 stations

4.1.5 Nutrients ($\text{NO}_2\text{-N}$, $\text{PO}_4\text{-P}$ and $\text{SiO}_3\text{-Si}$):

The highest concentration of $\text{NO}_2\text{-N}$ in Bashbaria station during monsoon and the value was $3.07 \mu\text{g/liter}$ and lowest value was $0.24 \mu\text{g/liter}$ during winter in the same station. The average value found in Bashbaria was $2.76 \mu\text{g/liter}$ (monsoon) and $0.29 \mu\text{g/liter}$ (winter), in Patenga was $1.73 \mu\text{g/liter}$ (monsoon) and $0.6 \mu\text{g/liter}$ (winter), in Teknaf was $0.64 \mu\text{g/liter}$ (monsoon) and $0.51 \mu\text{g/liter}$ (winter). A Two-way ANOVA results showed that variations in Nitrite-Nitrogen ($\text{NO}_2\text{-N}$) among 3 stations and 2 seasons were significant ($p < 0.05$).

$\text{PO}_4\text{-P}$ was found to vary within a wide range during the research. The highest value was $1.65 \mu\text{g/liter}$ at Teknaf station during winter in 10 meter depth and the lowest value was $0.32 \mu\text{g/liter}$ during monsoon in the same station. The average value found in Bashbaria was $0.55 \mu\text{g/liter}$ (monsoon) and $0.92 \mu\text{g/liter}$ (winter), in Patenga was

0.54 $\mu\text{g/liter}$ (monsoon) and 1.00 $\mu\text{g/liter}$ (winter), in Teknaf was 0.74 $\mu\text{g/liter}$ (monsoon) and 1.46 $\mu\text{g/liter}$ (winter). Two-way ANOVA results showed that variations in Phosphate-phosphorus ($\text{PO}_4\text{-P}$) among 2 seasons were significant ($p < 0.05$).

These nutrients were found increasing during monsoon rather than all seasons in the other sites except in Teknaf. Nevertheless, the highest value of $\text{SiO}_3\text{-Si}$ was 422.64 $\mu\text{g/liter}$ at Bashbaria during monsoon in surface layer and the lowest value was 55.52 $\mu\text{g/liter}$ at Patenga during winter in 5 meter depth. The average value found in Bashbaria was 231.02 $\mu\text{g/liter}$ (monsoon) and 141.48 $\mu\text{g/liter}$ (winter), in Patenga was 205.95 $\mu\text{g/liter}$ (monsoon) and 117.6 $\mu\text{g/liter}$ (winter), in Teknaf was 101.48 $\mu\text{g/liter}$ (monsoon) and 128.34 $\mu\text{g/liter}$ (winter). Two-way ANOVA results showed that variations in Silicate-Silicon ($\text{SiO}_3\text{-Si}$) among 3 stations and 2 seasons were significant ($p < 0.05$).

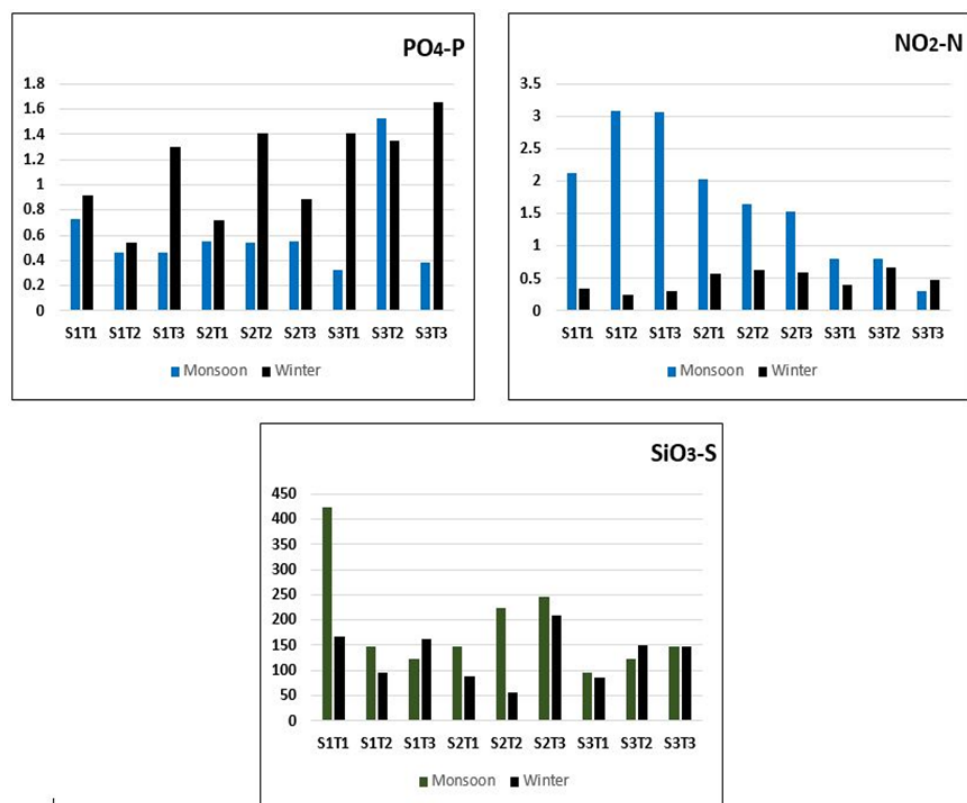


Fig. 12: Average Nutrients fluctuation among 3 stations

4.1.6 Chlorophyll-a:

As chlorophyll-a is an indicator of productivity, It was found higher in more productive Teknaf station during monsoon and this value was 1.61 $\mu\text{g/liter}$ in the

surface layer. The lowest value of chl-a was 0.12 $\mu\text{g/liter}$ in 10 meter depth during monsoon at Bashbaria station which considered as less productive region. The average value found in Bashbaria was 0.216 $\mu\text{g/liter}$ (monsoon) and 0.206 $\mu\text{g/liter}$ (winter), in Patenga was 0.686 $\mu\text{g/liter}$ (monsoon) and 0.463 $\mu\text{g/liter}$ (winter), in Teknaf was 1.396 $\mu\text{g/liter}$ (monsoon) and 1.123 $\mu\text{g/liter}$ (winter). Two-way ANOVA results showed that variations in Chlorophyll-a among 3 stations and 2 seasons and depth were significant ($p < 0.05$).

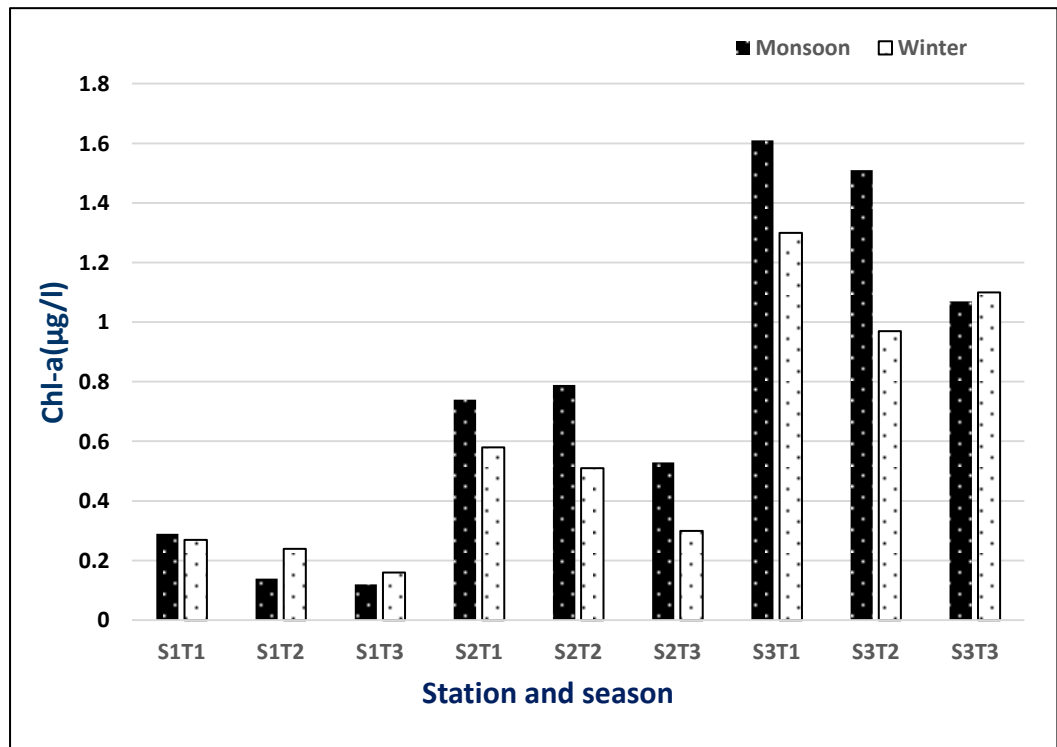


Fig. 13: Average Chl-a fluctuation among 3 stations

4.2 Phytoplankton Sinking Rate:

Phytoplankton sinking rate was calculated through SETCOL method during this study. Sinking rate varied from season to season, depth to depth and also from station to station. The highest sinking rate was observed as 3.1728 m day^{-1} at Patenga during winter at 5 meter depth and the lowest value was 1.3368 m day^{-1} at Bashbaria during winter at 10 meter depth. The average value found in Bashbaria was 2.6296 m day^{-1} (monsoon) and 1.8918 m day^{-1} (winter), in Patenga was 2.3088 m day^{-1} (monsoon) and 2.5608 m day^{-1} (winter), in Teknaf was 2.4042 m day^{-1} (monsoon) and 2.3339 m day^{-1} (winter). Two-way ANOVA results showed that variations in Phytoplankton Sinking Rate among 3 stations and 2 seasons and depth were significant ($p < 0.05$).

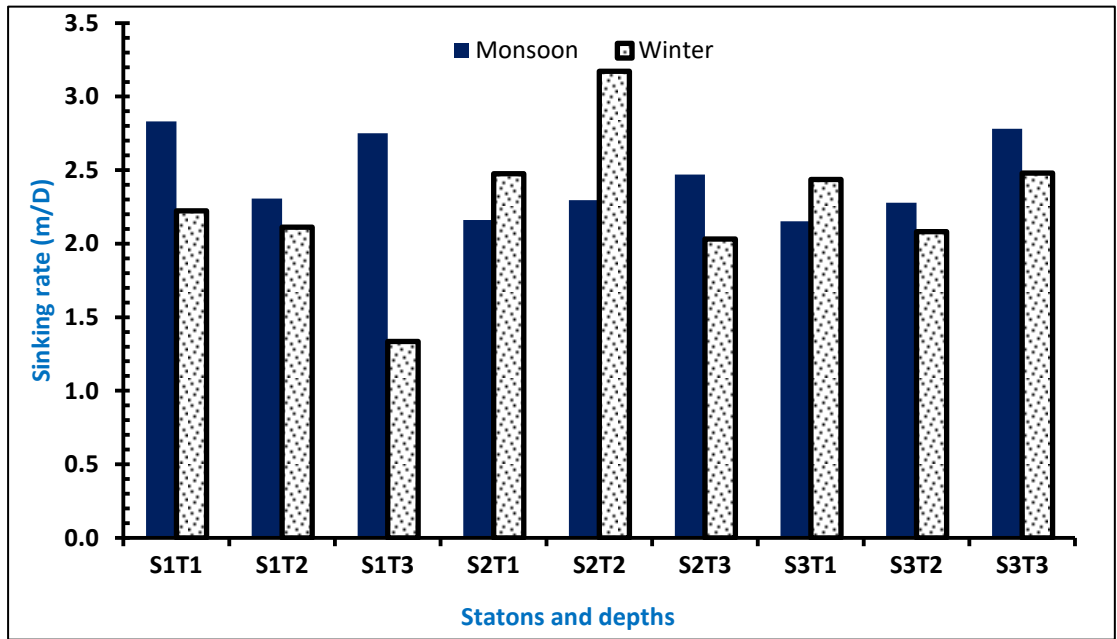


Fig. 14: Average Phytoplankton Sinking Rate fluctuation among 3 stations

4.3 Total Carbon:

During this study average total Carbon was varied from 2.166 - 2.111 mg/m³ in Bashbaria, 3.930 - 3.218 mg/m³ in Patenga and 7.986-6.298 mg/m³ in Teknaf during winter and monsoon respectively in the surface to 10 meter depth water column. Two-way ANOVA results showed that variations in Total Carbon among 3 stations and 2 seasons were significant (p<0.05).

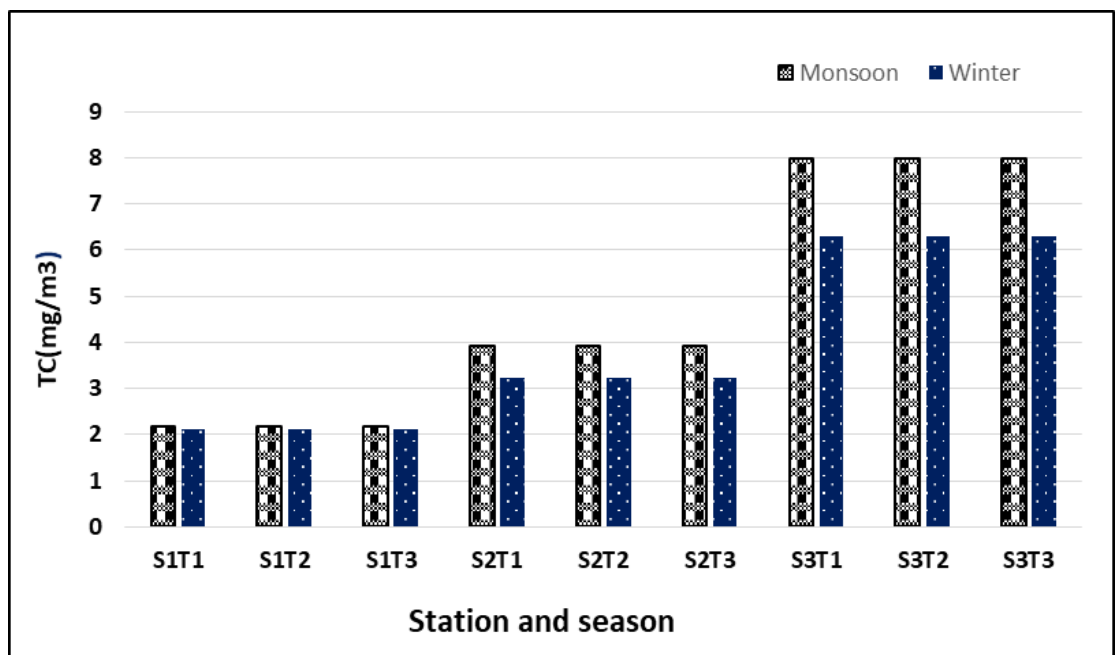


Fig. 15: Average TC fluctuation among 3 stations

4.4 Carbon flux:

During this study average Carbon flux varied from 5.69 ± 0.61 to 3.99 ± 1.02 mg C/m²d⁻¹ in Bashbaria, 9.07 ± 0.61 to 8.24 ± 1.84 mg C/m²d⁻¹ in Patenga and 19.20 ± 2.66 to 14.69 ± 1.37 mg C/m²d⁻¹ in Teknaf. Two-way ANOVA results showed that variations in Carbon flux among 3 stations and 2 seasons and depth were significant ($p < 0.05$).

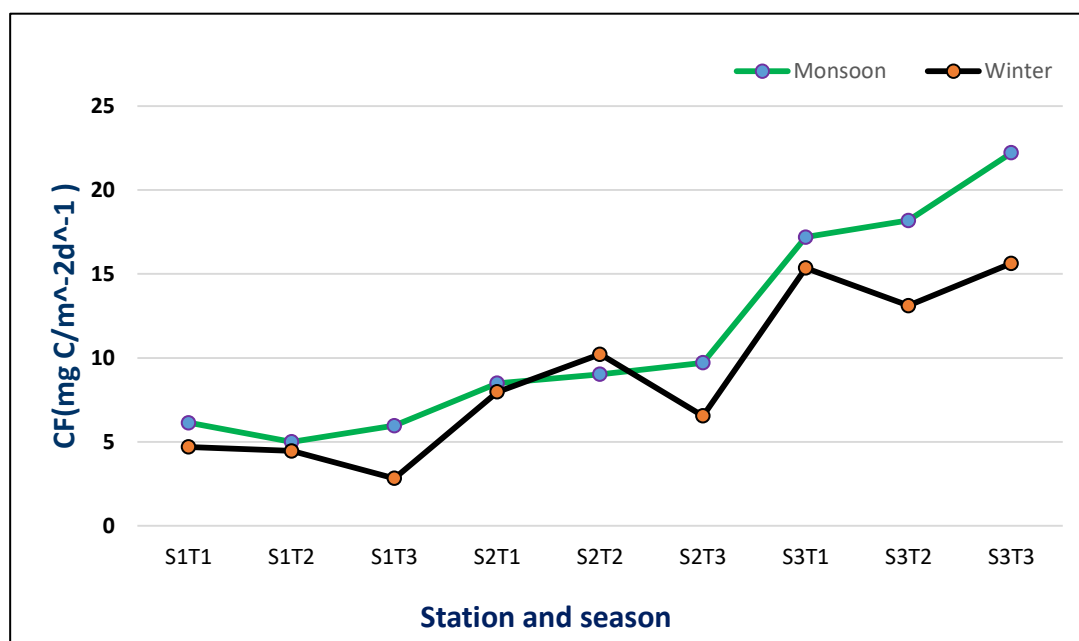


Fig. 16: Average Carbon Flux fluctuation among 3 stations

4.5 Phytoplankton composition and abundance:

Phytoplankton abundance and composition were recorded during this study period. The average value in Bashbaria was 5.2×10^2 cell/l (monsoon) and 4.9×10^2 cell/l (winter), in Patenga was 1.2×10^3 cell/l (monsoon) and 9.7×10^2 cell/l (winter), in Teknaf was 1.6×10^3 cell/l (monsoon) and 1.2×10^3 cell/l (winter). Three dominant class Bacillariophyceae, Dinophyceae and Chlorophyceae were found in these three stations. The average percentage of Bacillariophyceae was 87.5 % (monsoon) and 93.3% (winter) in Bashbaria, 85.8% (monsoon) and 90.2% (winter) found in Patenga and 85.1% (monsoon) and 82.8% (winter) found in Teknaf. Second dominant class of Dinophyceae was recorded 12.5% (monsoon) and 6.6% (winter) found in Bashbaria, 9.4 % (monsoon) and 5.0% (winter) found in Patenga and 12.8 % (monsoon) and 13.1 % (winter) in Teknaf. Among the three dominant classes Chlorophyceae was not in Bashbaria. The average percentage of Chlorophyceae 5.4% (monsoon) and 5.0 %

(winter) found in Patenga and 1.9% (monsoon) and 2.6 % (winter) found in Teknaf. Two-way ANOVA results showed that variations in phytoplankton abundance among 3 stations and 2 seasons were significant ($p < 0.05$).

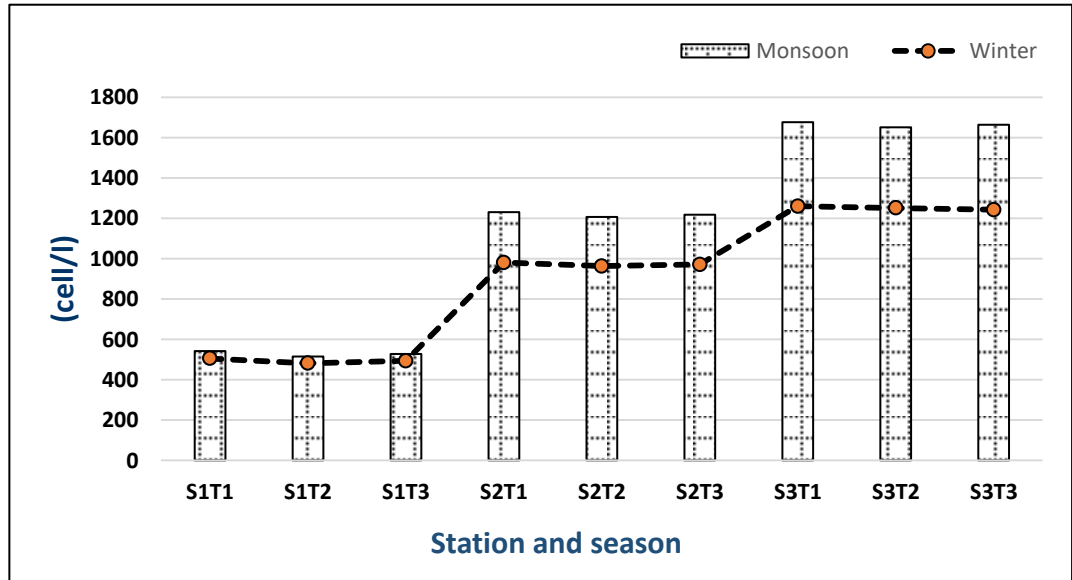


Fig. 17: Phytoplankton abundance fluctuation among 3 stations

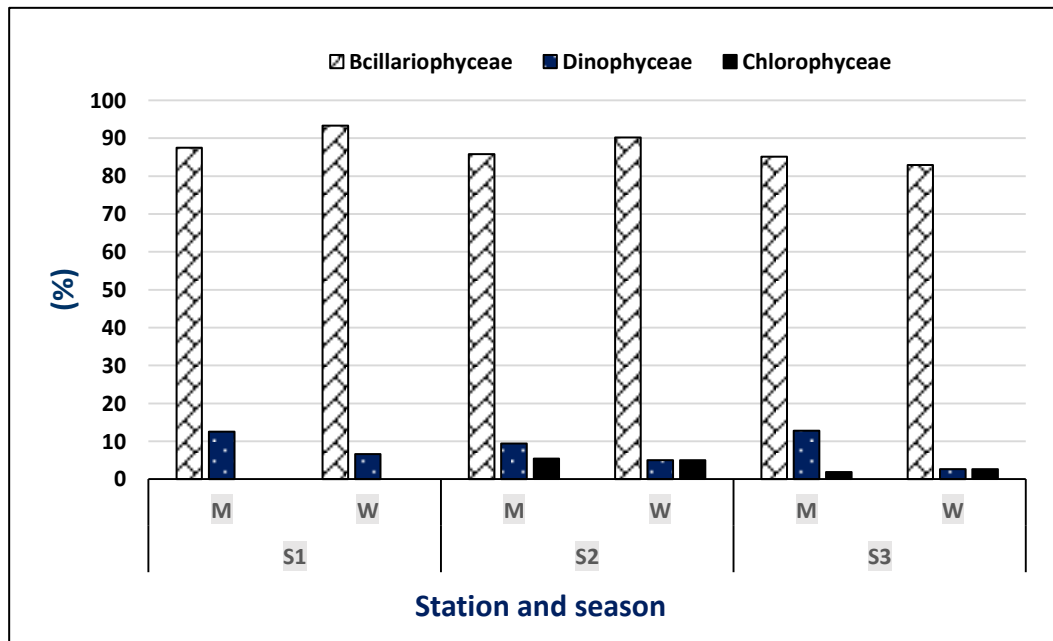


Fig. 18: Phytoplankton composition fluctuation among 3 stations

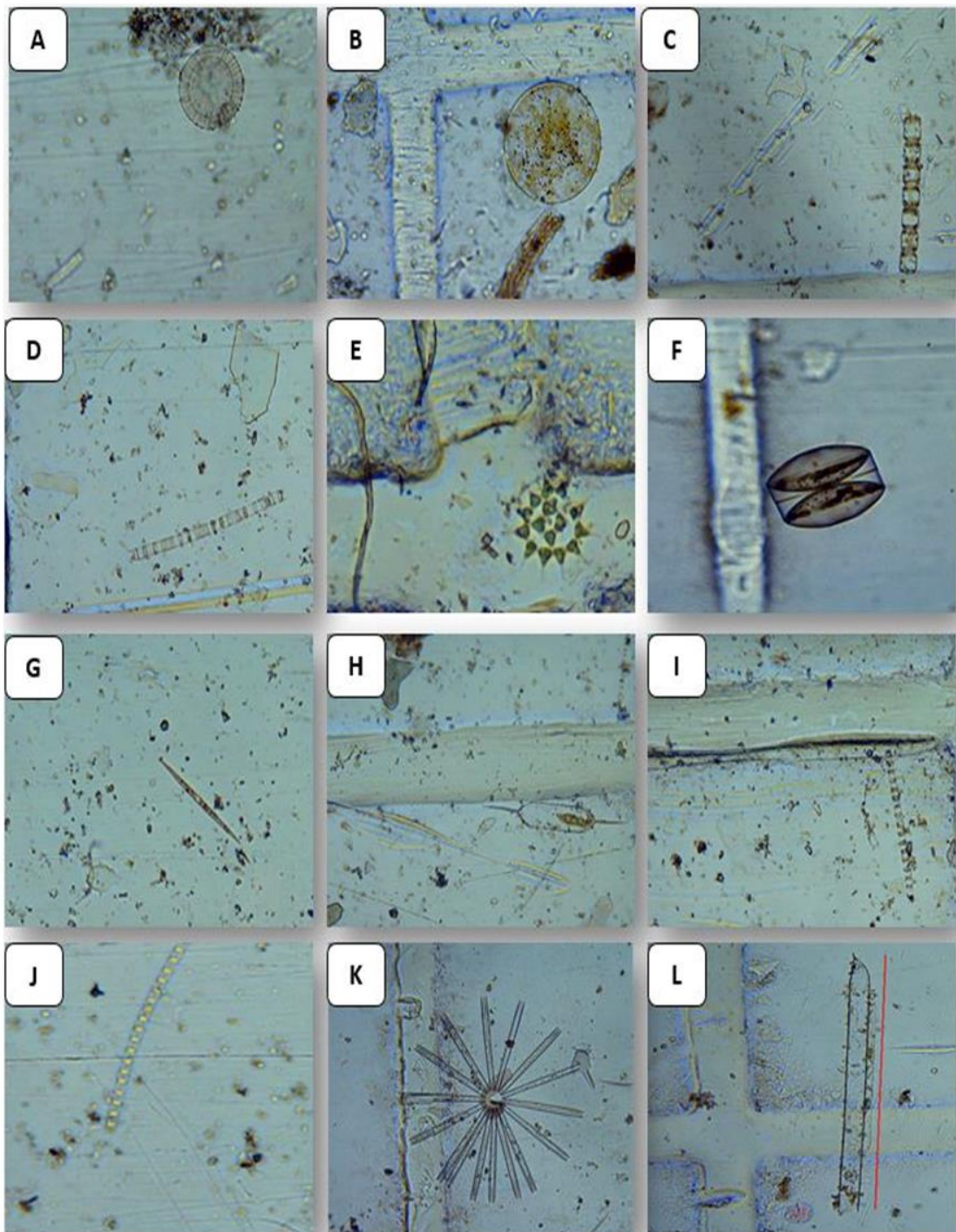


Fig.19 : Some phytoplankton found during study period A) *Cyclotell sp.*, B) *Coscinodiscus sp.*, C) *Lauderia sp.*, D) *Cerataulina sp.*, E) *Padiastrum sp.*, F) *Amphora sp.*, G) *Cyllindrotheca sp.*, H) *Dytillum sp.*, I) *Melosira sp.*, J) *Skeletonema sp.*, K) *Thalassiothrix sp.*, L) *Rhizosolenia sp.*

Parameters	BM	BW	PM	PW	TM	TW
WT (°C)	30.5±0.1	25.2±0.1	32.2±0.1	26.3±0.1	32.0±0.5	23.6±0.2
Salinity (psu)	10.1±0.2	16.5±0.5	3.6 ± 0.5	21.3±0.5	25.7 ± .2	32.6 ± .5
pH (ppt)	6.9 ± .02	7.5 ± .05	6.7 ± 0.1	7.1 ± .02	6.3 ± .05	6.5 ± .02
TDS (g/l)	11.1±.05	17.0±.05	2.8 ± .02	22.3±0.1	24.6 ± .9	30.5±0.1
TSS (g/l)	0.30±0.2	0.84 ± 0.07	0.29 ±0.2	0.94 ± 0.1	0.57 ±.48	0.31 ± .32
Chl-a (µg/liter)	0.18 ± .09	0.22 ± 0.05	0.68 ±.13	0.46 ± .14	1.39 ±.28	1.12 ± .16
NO ₂ -N (µg/l)	2.76 ± .54	0.29 ± 0.05	1.73 ±.25	0.6 ± 0.03	0.64 ±.28	0.51 ± .13
PO ₄ -P (µg/l)	0.55 ± .15	0.92 ± 0.38	0.54 ±.05	1.00 ± .35	0.74 ±.68	1.46 ± .15
SiO ₃ -Si (µg/l)	231±166	141.4 ±40.3	205.9±50	117.6±81	121.7±25	128.3 ±36
Sinking rate(m/d)	2.62 ± .28	1.89 ± 0.48	2.30 ±0.1	2.56 ± .57	2.22 ±.06	2.51 ± .09
Carbon flux(mg C/m ² ·d ⁻¹)	5.69 ± .61	3.99 ± 1.02	9.07 ±.61	8.24 ± .84	19.20 ± 2	14.69 ±1.3
Cell/l	521 ± 13	494 ± 12	1218 ±12	971.9 ±8.5	1663.2±1	1251.4 ± 9

Table 1: (Mean ± SD) value of three stations and two seasons (BM=Bashbaria Monsoon, BW=Bashbaria Winter, PM=Patenga Monsoon, PW=Patenga Winter, TM=Teknaf Monsoon and TW=Teknaf Winter)

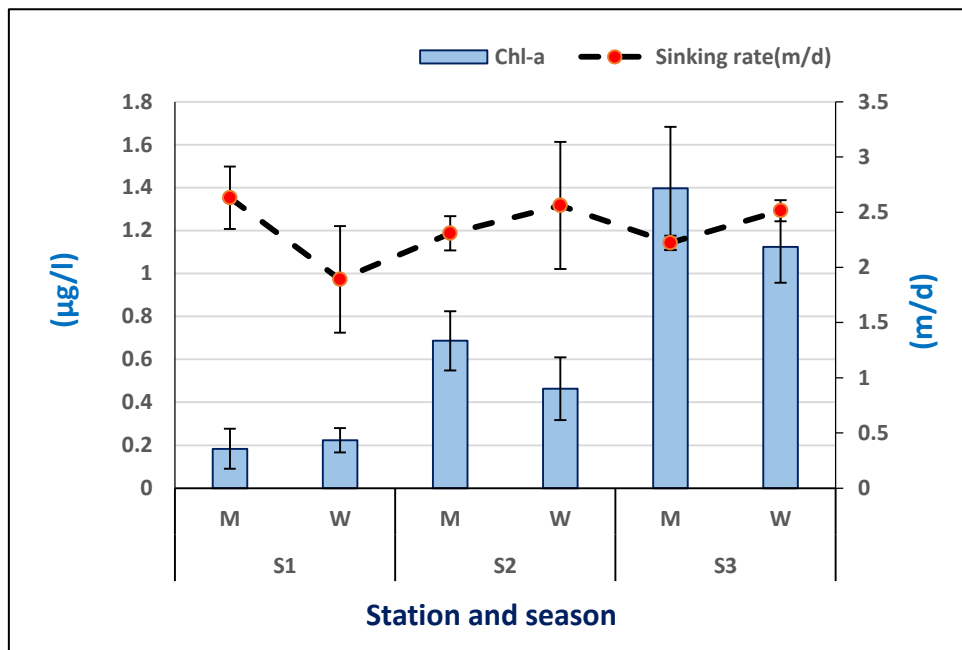
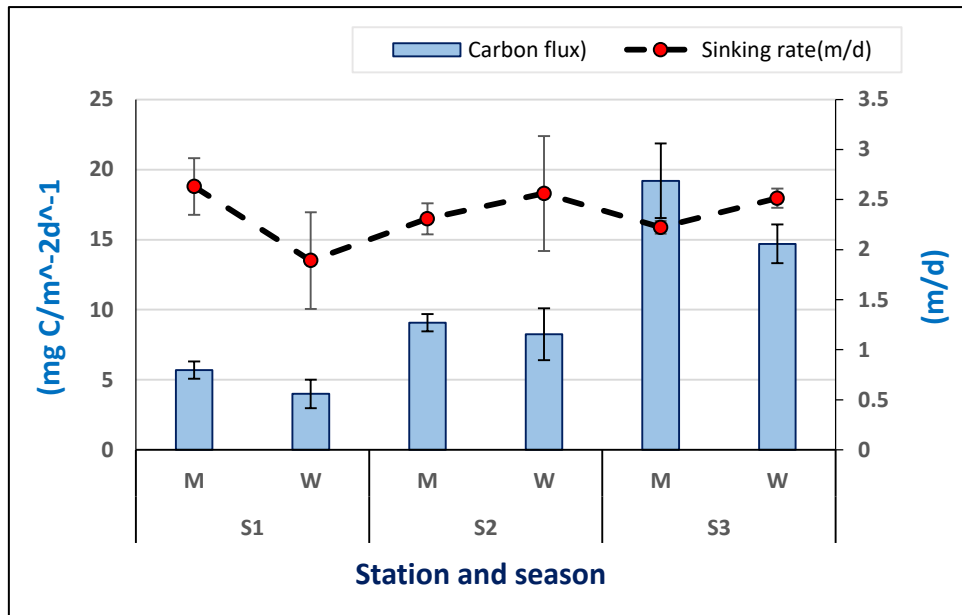


Fig.20: Average and SD value comparison between two factors.(A) Carbon flux and sinking rate (B) Chl-a and Sinking rate, (S1= Bashbaria,S2= Patenga,S3= Teknaf and M=Monsoon, W=Winter)

Table 2: Pearson correlation among the variables

Correlations													
	WT	pH	Salinity	Chl-a	TDS(g/L)	TSS(g/l)	NO ₂ -N	PO ₄ -P	SiO ₃ -Si	Sinking rate(m/d)	Total carbon(mg/m ³)	Carbon flux(mg C/m ² -2d ⁻¹)	cell/l
WT	1												
pH	.511*	1											
Salinity	.540*	.302*	1										
Chl-a	.243	.754*	.578**	1									
TDS(g/L)	.579*	-.230	.992**	.504**	1								
TSS(g/l)	.360*	.502*	.161	-.176	.229	1							
NO ₂ -N	.620*	-.222	.645**	-.320*	-.648**	-.493**	1						
PO ₄ -P	.628*	.090	.417**	-.024	.416**	.093	.354*	1					
SiO ₃ -Si	.275*	-.008	.425**	-.285*	-.427**	-.052	.318*	.001	1				
Sinking rate(m/d)	.063	.280*	.053	.052	.058	-.229	.283*	.145	.050	1			
Total carbon(mg/m ³)	.247	.793*	.632**	.953**	.564**	-.175	.323*	.035	.257	.009	1		
Carbon flux(mg C/m ² -2d ⁻¹)	.197	.820*	.660**	.936**	.591**	-.237	.291*	.051	.255	.214	.975**	1	
cell/l	.396*	.761*	.419**	.910**	.347*	-.159	-.261	.107	.217	.044	.932**	.910**	1

** . Correlation is significant at the 0.01 level (2-tailed).

* . Correlation is significant at the 0.05 level (2-tailed).

4.6.1 Depth wise PCA of physico-chemical parameters:

PC 1 and PC 2 combined were accounted for 71.8% of the total variance where Chl-a, CF and TC were showed highly correlation and formed cluster in 10 meter depth. TDS and salinity also showed correlation in this depth where Water Temperature had no direct correlation with other parameters.

On the other hand PC 2 and PC 3 were combinly accounted for 19.5% of the total variance where $\text{NO}_2\text{-N}$ and $\text{SiO}_3\text{-Si}$ correlated with 0 meter depth, $\text{PO}_4\text{-P}$ showed correlation with 5 meter depth and sinking rate with 10 meter depth. Among all factors WT, Chl-a and TC formed very close cluster.

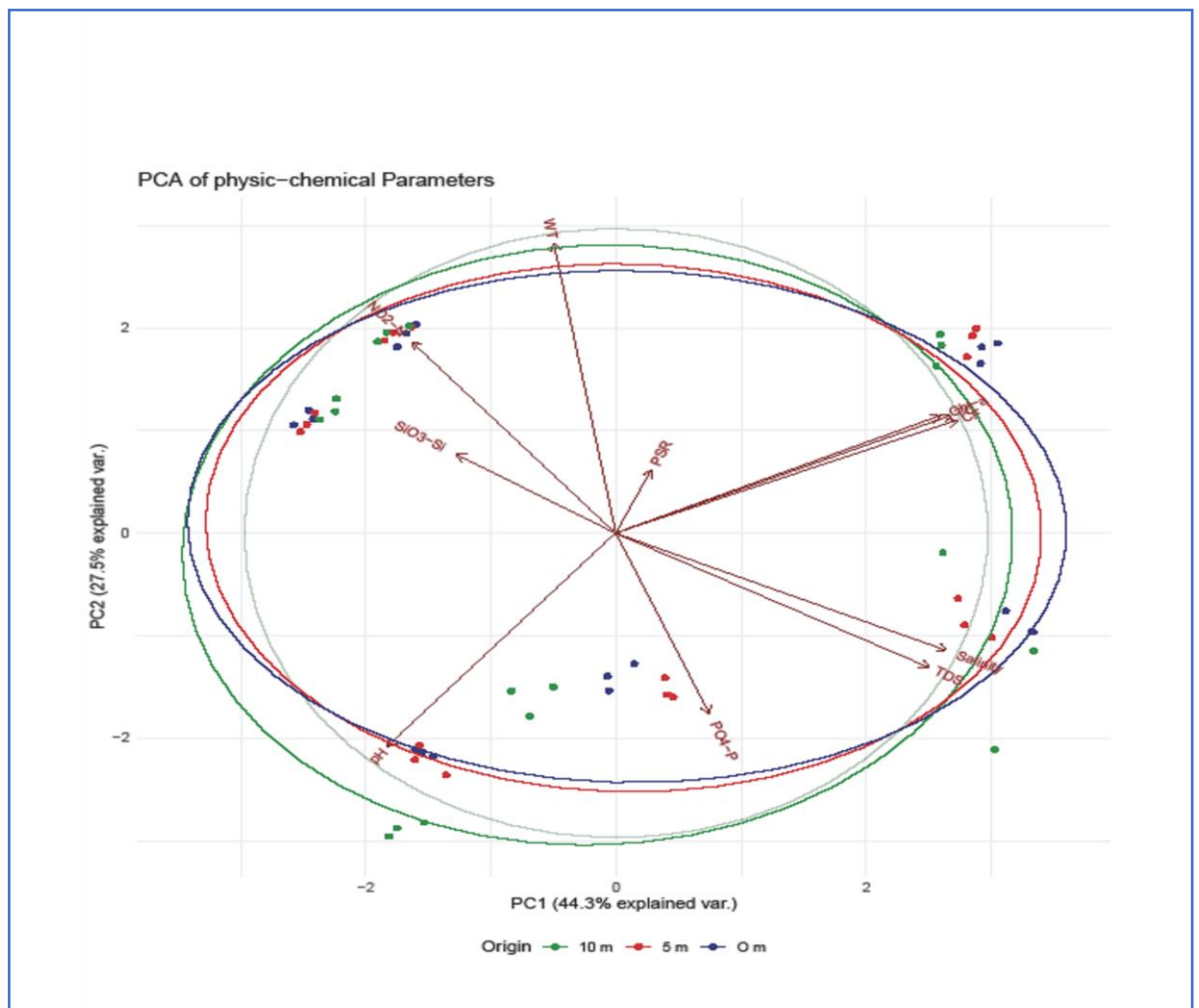


Fig. 21: PC 1 and PC 2 describing depth wise correlation of a particular station

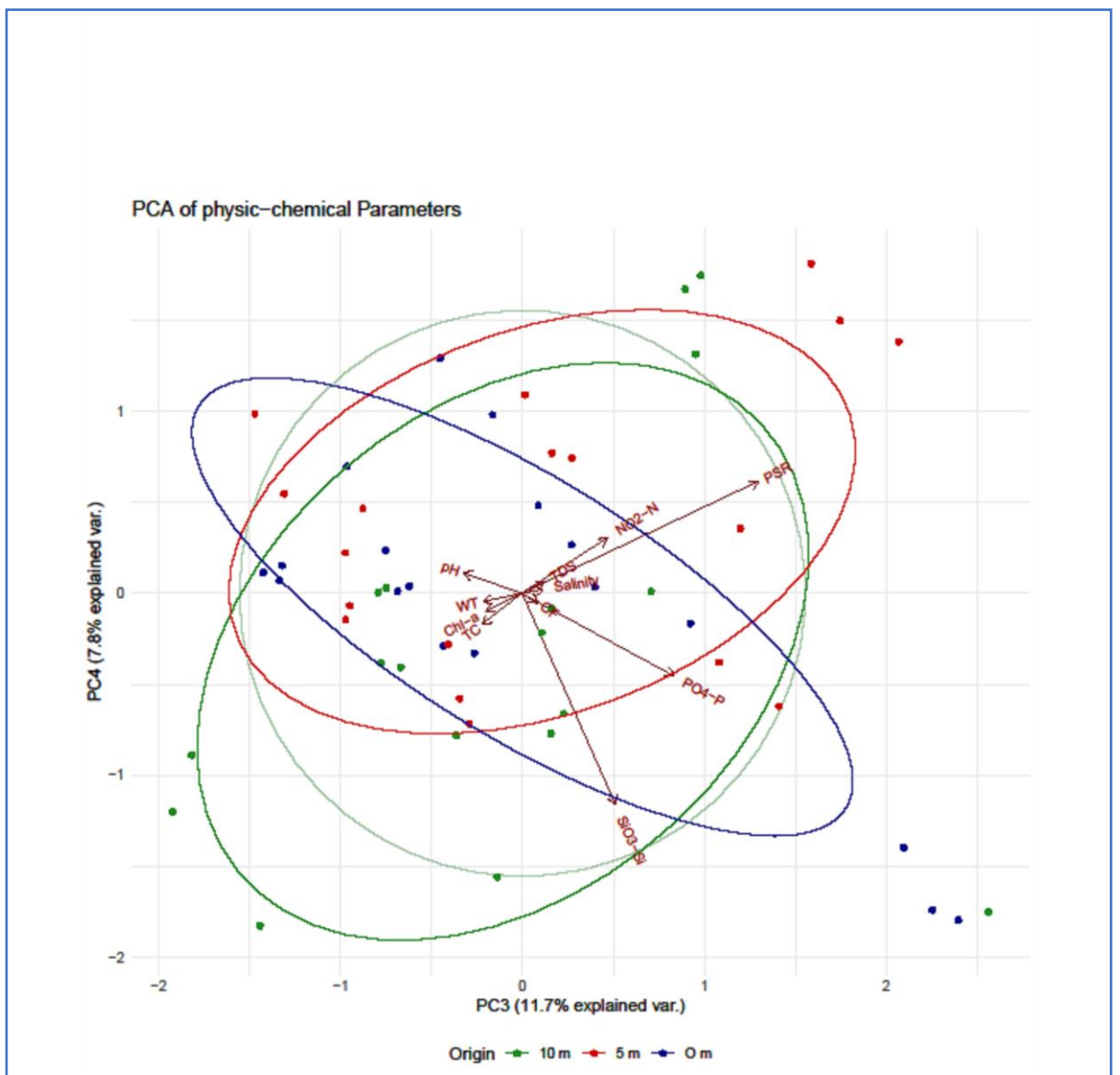


Fig. 22: PC 3 and PC 4 describing depth wise correlation of a particular station

4.6.2 Station wise PCA of physico-chemical parameters:

PC 1 and PC 2 combined were accounted for 71.8% of the total variance where Chl-a, CF and TC were showed highly correlated formed cluster in station 3. TDS and salinity also showed correlation in this station where pH had no correlation among three stations.

Both PC 3 and PC 4 combined were accounted for 19.5% of the total variance where sinking rate showed positive correlation with station 2.

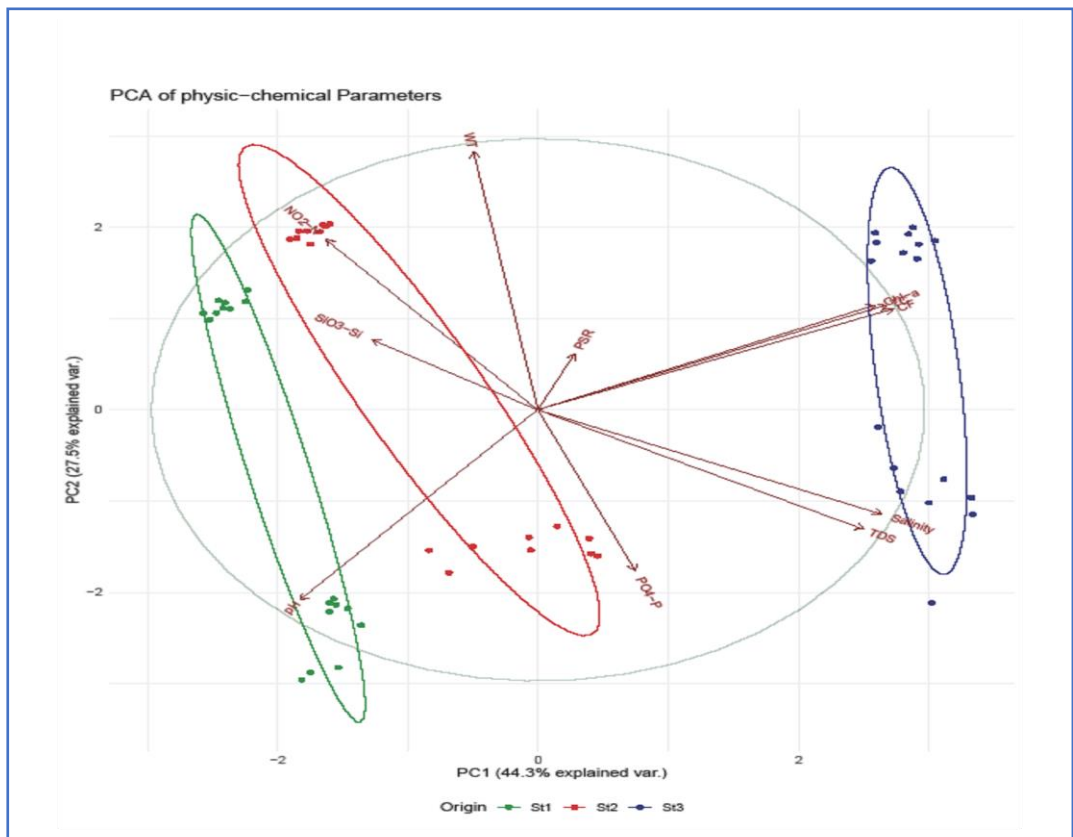


Fig. 23: PC 1 and PC 2 describing station wise correlation

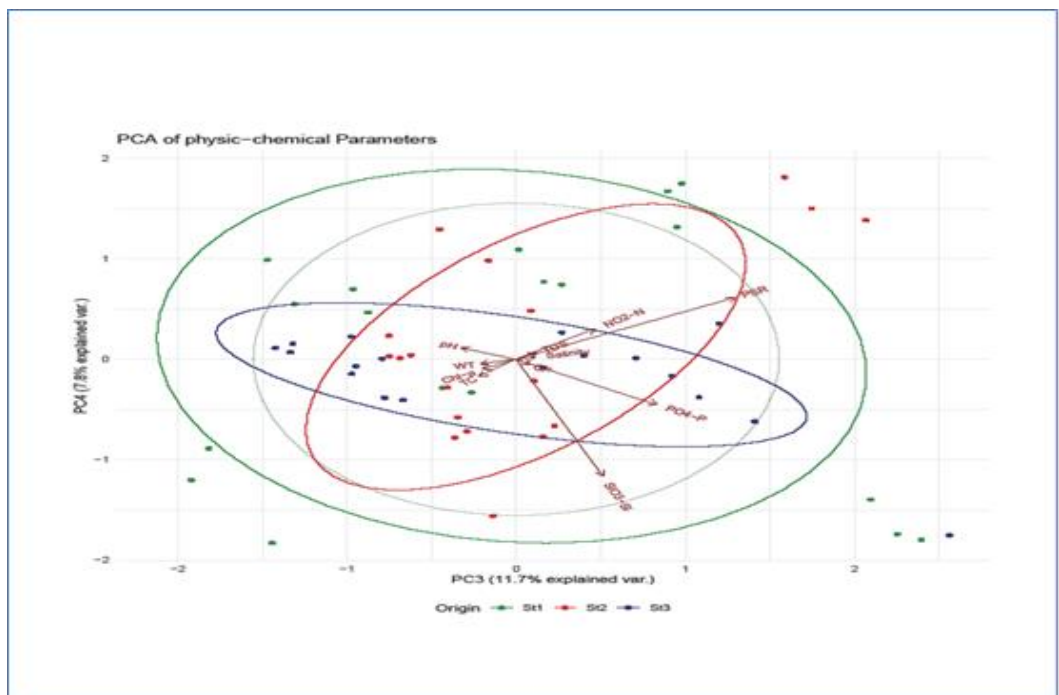


Fig. 24: PC 3 and PC 4 describing station wise correlation

4.6.3: Season wise PCA of physic-chemical parameters:

PC 1 and PC 2 combined were accounted for 71.8% of the total variance where salinity, TDS, pH and PO₄-P were showed positive correlation during winter and other two nutrients were positively correlated during monsoon. TC, CF and Chl-a had no seasonal correlation but strongly correlated among them.

On the other hand both PC 3 and PC 4 combined were accounted for 19.5% of the total variance where sinking rate showed positive correlation during monsoon and SiO₃-Si showed positive correlation in both season. And most of the rest parameters showed no correlation with season but showed correlation among them.

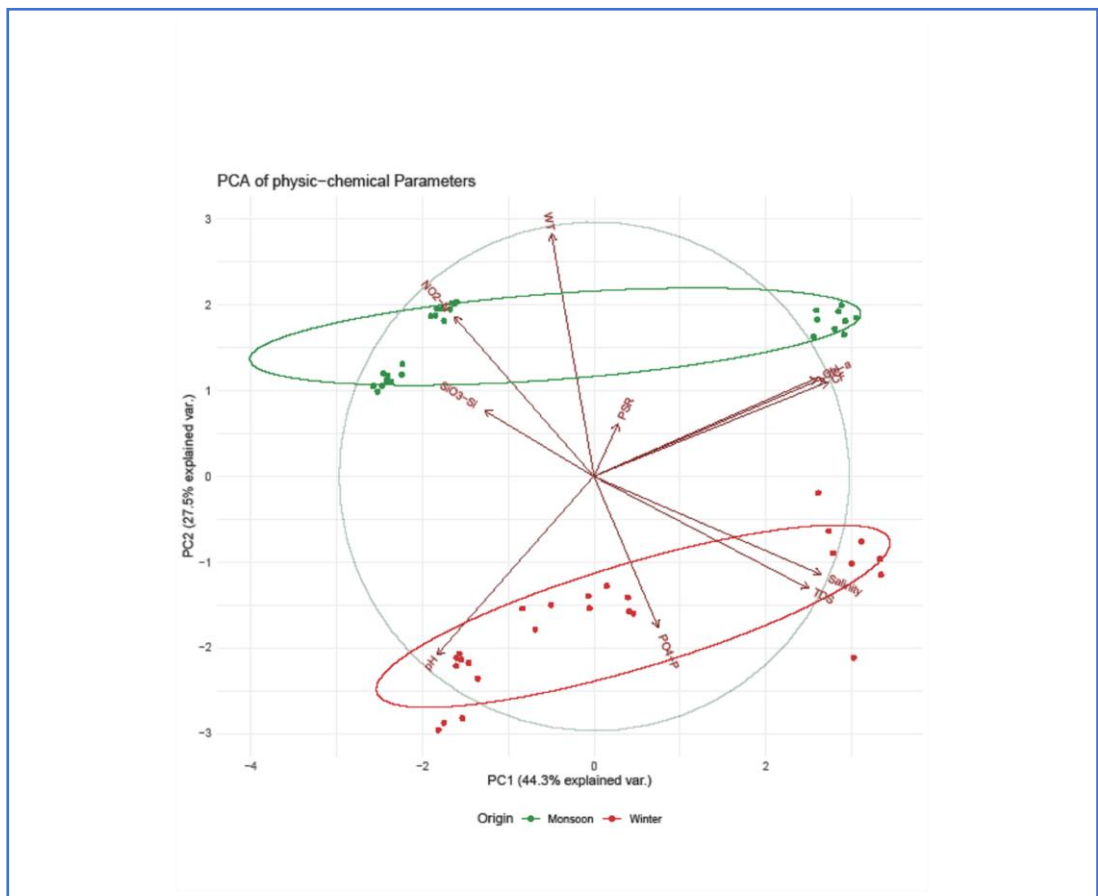


Fig. 25: PC 1 and PC 2 describing season wise correlation

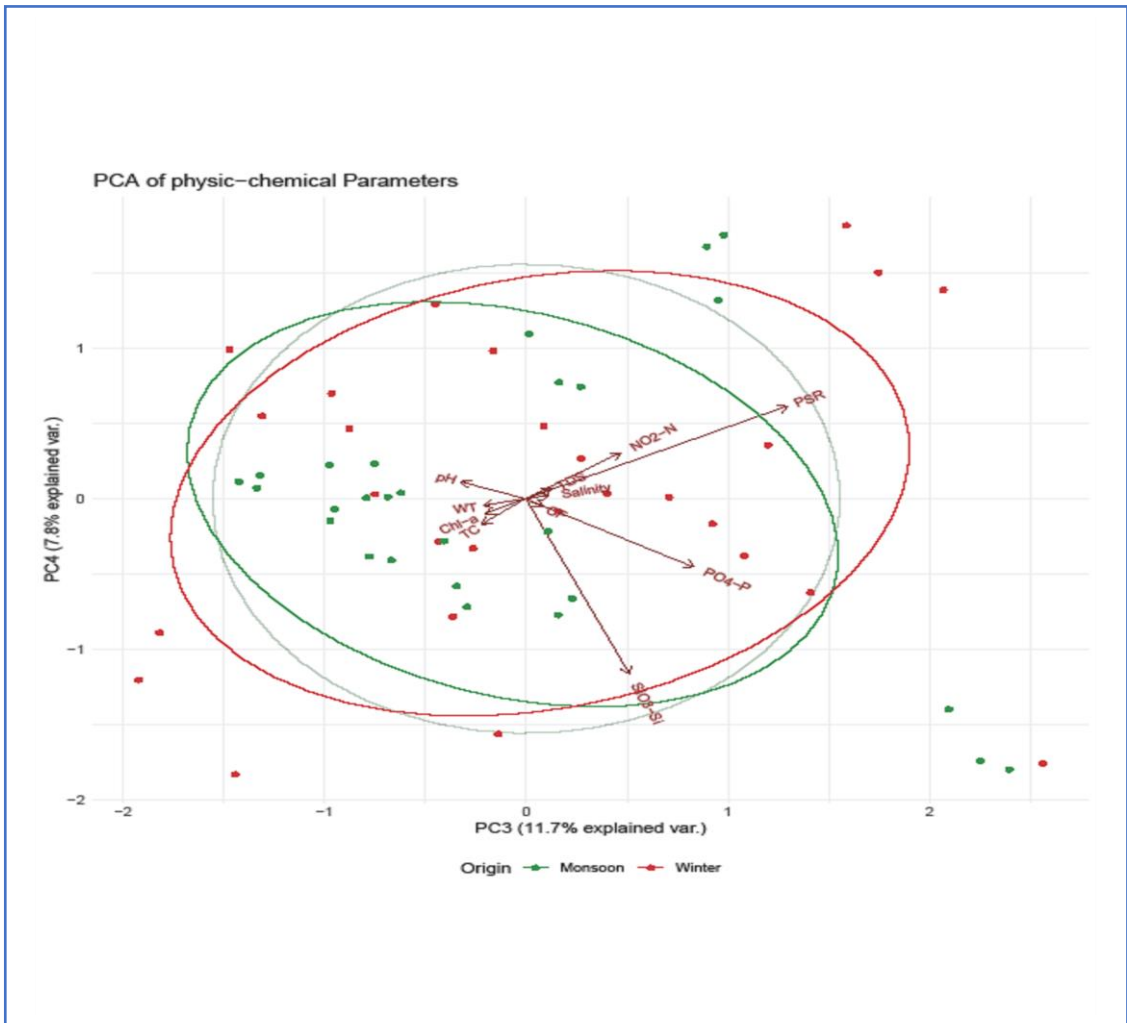


Fig. 26: PC 3 and PC 4 describing season wise correlation

CHAPTER FIVE

DISCUSSION

Ocean carbon sequestration through phytoplankton sinking rate associated with carbon flux from the surface to the bottom has been popular to the researcher after the invention of SETCOL method. This study was conducted at 3 selected stations of Northern Bay of Bengal belongs to Bangladeshi coast for the measurement of seasonal sinking of carbon with focusing on 10 dominant species of phytoplankton. There were both field and lab based analysis of parameters if they were correlated to sinking in the water column and the abundance of phytoplankton.

5.1 SETCOL method-it's applicability in field use:

Measurement of phytoplankton sinking rate in the ocean with various intensities of water turbulence was a great problem for the oceanographer until the invention of SETCOL method (Bienfang, 1981). Historically, There were several techniques used to determine phytoplankton sinking rate around the world including visual counting of cells with an inverted microscope (Smayda, 1965), fluorometric methods measuring in vivo chlorophyll a (Eppley *et al.*, 1967; Titman, 1975), detection of radioactively ¹⁴C labeled cells (Bienfang, 1979), optical density measurements using a spectrophotometer and SETCOL method (Bienfang, 1981). But except the radioactive and SETCOL method all other showed shortcoming in the measurement of sinking rate (Guo *et al.*, 2016). Due to complex procedure in radioactive method oceanographers prefer SETCOL method than the others. Because it is the only method to measure phytoplankton sinking rate without the disturbance of Turbulence. However, the sinking rate measured by SETCOL is an important parameter for understanding the motion of phytoplankton in seawater, regardless of turbulence.

5.2 Physico-chemical parameters:

Seasonal variation of water temperature among three stations was observed during this research. The observed range was higher in monsoon and lower in winter. Because temperature is relatively high during monsoon in north-east part of Bay of Bengal. Gwo-Ching Gong *et al.* (2002) found that temperature ranged from 13.0°C to 25.7°C in winter and 20.9 °C to 29.6 °C in summer in Bay of Bengal. But this research

showed that temperature increased toward monsoon from the winter due to ocean warming.

The average value of water pH found higher during winter and lower during monsoon. It showed that there was an inverse relationship between temperature and pH. Because molecular motions increased as the temperature increased (Matsumoto *et al.*, 2002) which resulted in water's tendency to ionize and form more hydrogen ions. As a consequence, the pH dropped. Mahmood (1976) studied in Karnofully estuary showed that water pH ranged from 7.10 to 7.30 during monsoon to winter.

There was found a direct relationship between high water discharge during monsoon and the water salinity. During this research the average value was lower during monsoon and higher during winter. The only reason behind the lower salinity during monsoon due to add of land run off and rainfall which diluted the water and decreased of salinity. The lowest value was recorded at Patenga during monsoon because the station was very adjacent to Karnofully estuary mouth and while monsoon heavy freshwater added after rainfall. But tekna station was not situated in the mouth of estuary like patenga so there was lower inland discharge than the patenga.

Ahmed (1989) found that 3.57 to 24.10 psu during January and August studied near the Chittagong coast.

According to Parvez *et al.* (2018) TDS Value along the ship breaking area, Chittagong coast ranged from 4.5 to 17.0 g/l during August and January respectively. In this research the average TDS value was higher during winter than monsoon. There was a positive correlation between them and showed higher value during winter than monsoon in every stations. On the other hand average TSS value found in Bashbaria was 0.30 g/l (monsoon) and 0.84 g /l (winter), in Patenga was 0.29 g/l (monsoon) and 0.94 g/l (winter), in Teknaf was 0.57 g/l (monsoon) and 0.31 g/l (winter).

Three nutrient components were measured to observe a relationship with other parameters and mostly with phytoplankton sinking rate and carbon flux between two seasons. It showed significant variation among stations and between seasons. The average value of Nitrite Nitrogen (NO₂-N) was higher in both Bashbaria and patenga station than the Teknaf. According to Noori (1999) the average value was varied from 0.126 to 1.198 µg/liter during monsoon in the south East coast of Bangladesh (Bay of

Bengal). But in this research higher value was found both in Bashbaria and Patenga due to heavy run-off and growing industries.

Noori (1999) also found the average value of Phosphate-phosphorus ($\text{PO}_4\text{-P}$) was 0.410 to 2.330 $\mu\text{g/liter}$ during this season whereas this study showed that during monsoon found higher value in both Potenga and Bashbaria due to high run-off but inverse in Teknaf.

5.3 Phytoplankton abundance and parameters effect:

As phytoplankton was the key indicator of carbon flux, it was recorded abundance station and season wise. Study showed that phytoplankton abundance was lower in winter in every station because monsoon provided huge run-off with nutrients. But due to highly turbid water the Bashbaria station was very poor in phytoplankton abundance though nutrients were high. And Teknaf station was showed high abundance with species variation due to transparent water. Diatom percentage were 87.7%, 85.81% and 85.14% in Bashbaria, Patenga and Teknaf respectively which indicated that the coast line was dominant by diatom species. According to Guo *et al.* (2016) diatom was the dominant species. Mehedi *et al* (2017) showed that the density of the phytoplankton cells ranged between 9408 and 21,964 cells L^{-1} in Reju khal estuary. There was difference between this study and the cited paper above because of sample collection method. They took sample by horizontally towing and in this paper vertical sample collection was done.

5.4 Phytoplankton sinking rate and associated factors:

During this study SETCOL method was used to determine phytoplankton sinking rate through chl-a measurement. Phytoplankton sinking rates ranged from 0.13 to 1.04 m day^{-1} in spring and 0.28 to 1.71 m day^{-1} in summer (Guo *et al.*, 2016) with two dominant species *Prorocentrum dentatum* and *Skeletonema dorhnii*. During the two cruises, with the exception of temperature and nitrite concentration in summer, no substantial correlation was observed between phytoplankton sinking rates and most environmental parameters. Analysis showed significant variation ($p < 0.05$) against depth, season and station and weakly correlated with most of the environmental parameters. A strong correlation was observed between the sinking rates of phytoplankton and the community structure of phytoplankton (Guo *et al.*, 2015). Another study showed that the sinking rates were mainly determined by the density of

cells or colonies rather than their size (Peperzak *et al.*, 2003). During monsoon the percentage of diatom abundance was the major contribution of total phytoplankton. According to Guo *et al.* (2016) when diatoms (dominant species) were overwhelming the population in summer, the sinking rates were relatively higher. The most possible reason why sinking rate of diatom was higher than the increased density resulting from silicification caused fast sinking (Raven, 2004). In this research Diatom percentage were 87.7%, 85.81% and 85.14% in Bashbaria, Patenga and Teknaf respectively. During winter this percentage was higher than the monsoon at all stations and it was considered as the most nearest reason why the sinking rate was higher in winter than monsoon in this station. The rate of sinking depends on the turbulence level and the simple dependence of sinking speed on radius can be altered by turbulence (Provenzale, 2010) and for this reason during high turbulent monsoon phytoplankton sinking rate was higher than winter in this study. The transparent exopolymer particles (TEP)—several studies confirmed that the associated dynamics of aggregation played an important part in the sedimentation of *Skeletonema sp.* blooms. It also regarded as a great factor in sinking of phytoplankton. But there is no plenty data related to it and need to further research.

5.5 Carbon flux:

Daily carbon flux showed the gradient of the values among depth, stations and seasons. Average value ranged from 3.6867- 5.5939 ($\text{mg C m}^{-2} \text{ day}^{-1}$) in Bashbaria, 7.9922- 8.2530 ($\text{mg C m}^{-2} \text{ day}^{-1}$) in Patenga and 14.6993-18.7070 ($\text{mg C m}^{-2} \text{ day}^{-1}$) in Teknaf during winter to monsoon respectively. In all stations the value was higher during monsoon. The most reliable reason of it was due to the abundance of phytoplankton and sinking rate. During monsoon the abundance of dominant species and their total cell carbon in water column were higher than the winter. As a result during monsoon carbon flux from surface to bottom was more than 1 fold higher than the winter. Carbon flux was 2.4 folds higher in summer than the spring during survey by Guo *et al.* (2016) in the Changjiang (Yangtze River) estuary, China. During the spring *P. dentatum* bloom they found phytoplankton carbon flux ranged from 9.29 $\text{mg C m}^{-2} \text{ day}^{-1}$ to 82.44 $\text{mg C m}^{-2} \text{ day}^{-1}$ and it proved that due to phytoplankton bloom carbon flux was high. But in this study there was no bloom and in Bashbaria and Patenga dominant species abundance was relatively low so carbon flux was comparatively lower than the above sited paper.

5.6 PCA discussion among depth, station and season:

Chl-a, CF and TC were showed positive correlation in 10 meter depth (PC1 and PC2) and NO₂-N and SiO₃-Si correlated with 0 meter depth, PO₄-P showed correlation with 5 meter depth and sinking rate with 10 meter depth. Among three stations Teknaf showed highly positive correlation within Chl-a, CF and TC and Patenga showed highly positive correlation with sinking rate. Season-wise PCA showed that salinity, TDS, pH and PO₄-P had positive correlation during winter and other two nutrients were positively correlated during monsoon. TC, CF and Chl-a had no seasonal correlation but strongly correlated among them.

After evaluating the effect of changing sinking rates on phytoplankton dynamics and associate carbon flux in the northern Bay of Bengal, particularly in the south-eastern shallow coastal region of Bangladesh, it is clearly said that among three stations Teknaf coastal region contribute higher carbon absorption daily than other two region and obviously it is higher in monsoon than winter.

CHAPTER SIX

CONCLUSION

Almost half of the earth carbon is absorbed by the ocean's primary producer through "Biological pump". Phytoplankton plays not only as a primary producer but also as a vehicle of carbon export from the surface to the depth of ocean. So, most importantly sinking of phytoplankton and phytoplankton abundance determine the rate of daily carbon flux. Carbon absorption may vary from season to season and higher carbon flux also indicates about the higher productivity of this specific region. The study was conducted in three major points of Northern east coast of Bay of Bengal. It included three depth of water column to understand the difference of carbon sinking among depth. Phytoplankton sinking rate was higher in winter than monsoon in both Potenga and Teknaf station except Bashbaria station. Carbon flux was higher in Teknaf station and lower in Bashbaria. Pearson correlation showed that there was no significant correlation between phytoplankton sinking rate and most of the parameters except Nitrite.

From this study it can be said that Teknaf station is the highest contributor of daily carbon absorption than another two stations. During this research only two major season was covered due to lack of enough fund and facilities and only three stations of north-east part of Bay of Bengal were selected. But the north-east coast of Bay of Bengal belongs to Bangladesh about 711 km long. So, the next step should be done this research in the rest part of the coast throughout the year. But it is matter of hope that through this study a lots of data about carbon sinking can be recorded successfully and it will be the pioneer research for further study in this same field in another coast. Methods that are used in this study will be very helpful to upcoming researcher to get accurate value to rich their research work. Finally these data will give concept about the yearly carbon absorption in whole coastal area of Bangladesh through further research and be helpful in making national rules and regulations.

CHAPTER SEVEN

RECOMMENDATIONS AND FUTURE PERSPECTIVES

Research is the investigation of a particular topic to find accurate data to get a clear concept on this subject. During this research seasonal carbon absorption and phytoplankton sinking rate as well as impact of physico-chemical parameters on daily sinking were studied. It was found that comparatively highly productive water body contributes more in carbon absorption than the less productive water bodies and Bashbaria station was lowest and Teknaf was highest contributor in case of daily carbon sinking. As this research is still the first work in the field of carbon sinking of Bangladeshi coastal area, it can be done in other coast of the country. Bangladesh has a 711 km long coastal area and so unstudied region should be studied. Besides we have many freshwater water bodies like river, lake, haor, baor etc. As atmospheric carbon is a key indicator of environmental status, this research should be done in these freshwater bodies also. Though this work is mainly ocean based and risky also, so well decorated research vessel should be included to get more accurate data and make sure the safety. Updated methods could be included also to expand research interest. Hopefully this research can be used as secondary data source which will help future researcher in home and abroad and also help the national law maker in environmental issues about ocean carbon.

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APPENDICES

A. Table of pairwise comparison of sinking rate

Pairwise Comparisons

Dependent Variable: Sinking rate(m/d)

(I) Depth	(J) Depth	Mean Difference (I-J)	Std. Error	Sig. ^b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
0	5	-.083 [*]	.001	.000	-.085	-.082
	10	.163 [*]	.001	.000	.161	.165
5	0	.083 [*]	.001	.000	.082	.085
	10	.247 [*]	.001	.000	.245	.249
10	0	-.163 [*]	.001	.000	-.165	-.161
	5	-.247 [*]	.001	.000	-.249	-.245

Based on estimated marginal means

*. The mean difference is significant at the 0.05 level.

b. Adjustment for multiple comparisons: Least Significant Difference (equivalent to no adjustments).

Pairwise Comparisons

Dependent Variable: Sinking rate(m/d)

(I) Season	(J) Season	Mean Difference (I-J)	Std. Error	Sig. ^b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
Monsoon	Winter	.065 [*]	.001	.000	.064	.067
Winter	Monsoon	-.065 [*]	.001	.000	-.067	-.064

Based on estimated marginal means

*. The mean difference is significant at the 0.05 level.

b. Adjustment for multiple comparisons: Least Significant Difference (equivalent to no adjustments).

Pairwise Comparisons

Dependent Variable: Sinking rate(m/d)

(I) Station	(J) Station	Mean Difference (I-J)	Std. Error	Sig. ^b	95% Confidence Interval for Difference ^b	
					Lower Bound	Upper Bound
BASHBARIA	PATENGA	-.174 [*]	.001	.000	-.176	-.172
	TEKNAF	-.107 [*]	.001	.000	-.109	-.105
PATENGA	BASHBARIA	.174 [*]	.001	.000	.172	.176
	TEKNAF	.067 [*]	.001	.000	.065	.069
TEKNAF	BASHBARIA	.107 [*]	.001	.000	.105	.109
	PATENGA	-.067 [*]	.001	.000	-.069	-.065

Based on estimated marginal means

*. The mean difference is significant at the 0.05 level.

b. Adjustment for multiple comparisons: Least Significant Difference (equivalent to no adjustments).

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Appendix B: A book of cell volume measurement

Brief Biography of the author

This is Mishu Acharjee; son of Poresh Acharjee and Subarna Acharjee from Eidgah thana under the district of Bangladesh. He passed the Secondary School Certificate Examination in 2011 from Eidgah Model High School and Higher Secondary Certificate Examination in 2013 from Omar Gani M.E.S College, Chattogram. He obtained his B. Sc. in Fisheries (Hons.) Degree in 2018 from Faculty of Fisheries, Chittagong Veterinary and Animal Sciences University (CVASU), Chattogram, Bangladesh. Now, he is a candidate for the degree of MS in Marine Bioresource Science under the Department of Marine Bioresource Science, Faculty of Fisheries, CVASU. He has great interest on scientific research on Marine Science.