

Study on Diversity and Abundance of Phytoplankton in Coastal Waters of Goa

A Dissertation for

Course code and Course Title: MMD 412 Dissertation

Credits: 8

Submitted in partial fulfilment of Masters Degree

M.Sc.in Marine Microbiology

by

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Date: **APRIL 2023**

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DECLARATION BY STUDENT

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I hereby declare that the data presented in this Dissertation report entitled, “Study on diversity and abundance of phytoplankton in coastal waters of Goa” is based on the results of investigations carried out by me in the M.Sc. in Marine Microbiology at the School of Earth, Ocean and Atmospheric Sciences, Goa University under the Supervision/Mentorship of Dr.Varada S. Damare and the same has not been submitted elsewhere for the award of a degree or diploma by me. Further, I understand that Goa University or its authorities will be not be responsible for the correctness of observations / experimental or other findings given the dissertation.

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This is to certify that the dissertation report "**Study on Diversity and Abundance of Phytoplankton in Coastal Waters of Goa**" is a bonafide work carried out by **Ms Nitisha Nilesh Gaonker** under my supervision in partial fulfilment of the requirements for the award of the degree of **Masters** in the Discipline Marine Microbiology at the (School of Earth, Ocean and Atmospheric Sciences), Goa University.

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ACKNOWLEDGEMENT

I would like to express my deep sense of gratitude and sincere thanks to my beloved guide Dr. Varada S. Damare, Assistant Professor, Marine Microbiology, Goa University; for her guidance, patience and for helping me throughout with the dissertation and calmly solving all my queries.

I am grateful to Dr. Priya M. D'Costa, Programme Director of Marine Microbiology for helping me to identify the phytoplankton species for my dissertation work without whom this project would not have been a success.

I would also like to express my heartfelt thanks to all the teachers of the Department of Marine Microbiology for their time and help rendered, and all the non teaching staff of the department for their help and support. I would also like to thank our lab assistant Ms Vaishali Merchant for her support and help in the laboratory.

A wholehearted thank you to all my loving friends and my classmates for their love and support and for giving a constant motivation throughout the work.

Last but not the least, a big thank you to my wonderful parents and my family who have always motivated me to achieve my goals.

INDEX

SR. NO.	CHAPTERS	PAGE NO.
1	INTRODUCTION	1-14

2	MATERIALS AND METHODS 2.1 STUDY SITE AND SAMPLING STRATEGIES 2.2 PHYSICO-CHEMICAL ANALYSIS 2.3 ANALYSIS OF PHYTOPLANKTON SAMPLES	15-20
3	OBSERVATION	21-45
4	RESULT AND DISCUSSION	46-53
5	SUMMARY	54
6	BIBLIOGRAPHY	55-60

TABLE NO.	LIST OF TABLES	PAGE NO.
1	Location of sampling sites	16
2	Phytoplankton recorded in Anjuna sample	22

3	Phytoplankton recorded in Caranzalem sample	28
4	Phytoplankton recorded in Dona Paula sample	32
5	Phytoplankton recorded in Miramar sample	36
6	Phytoplankton recorded in Siridao sample	41
7	Phytoplankton recorded in MandoviZuari sample	44
8	Abundance of phytoplankton cells/L	49
9	Species observed in samples	51

SR. NO.	FIGURE NO.	PAGE NO.
1	Types of phytoplankton	3
2	Schematic phylogenetic tree representing the distribution of phytoplanktonic taxa across eukaryote lineages	4

3	The structure of a diatom cell	5
4	Conceptual diagram of life cycle of the centric diatoms	6
5	A variety of dinoflagellates from the NE Atlantic	7
6	General structure of Dinoflagellate	8
7	Conceptual overview of the dinoflagellate life cycle	9
8	Geographic location of water collection sites	16
9	Lugol's solution	18
10	Siphoned samples	18
11	Sedgewick Rafter cell	20
12	Salinity of water samples from all the sampling locations	48
13	pH of water samples from all the sampling locations	49

CHAPTER 1

INTRODUCTION

INTRODUCTION

Marine phytoplankton, also known as the autotrophic part of the plankton (from the Greek words "phyton" for plant and "planktos" for wanderer), are organisms that live in the ocean's well-lit surface layers, down to a depth of 200 m in the clearest waters, and use photosynthesis to produce energy. The majority of phytoplankton species are microscopic, unicellular organisms (**Simon et al., 2009**)(**Figure 1**). Not only are phytoplankton cells taxonomically varied, but they also have large size range within the groups of taxa. Even the size spectra might alter over a period of time and/or space as a result of shifting environmental factors or succession of life cycle stages. There are more than three orders of magnitude between the size of phytoplankton cells, which range from picoplankton (0.2-2 μm) to mesoplankton (0.2– 2 mm)(**Not et al.,2012**).

Due to large range of cell sizes, various collecting mechanisms such as plankton nets and filters on various mesh sizes, as well as observation approaches (optical and infrared), must be deployed to investigate phytoplankton diversity through electron microscopy (**Not et al.,2012**).

More than 45% of the annual net primary production of our planet is produced by marine phytoplankton. In comparison to the biomass of terrestrial photosynthetic species, the biomass is kept extremely low through continuous grazing by zooplankton and recycling (**Simon et al., 2009**).

Most other aquatic organisms are supported by marine phytoplankton, which supports essential global processes such as oxygen synthesis, carbon fixation and CO_2 sequestration, as well as nutrient cycling. Diatoms, dinoflagellates, haptophytes, and small prasinophytes are the most diverse and environmentally relevant eukaryotic phytoplankton groups in current oceans. These species are also responsible for the creation of the massive blooms visible in satellite images (**Not et al.,2012**).



Figure 1. Phytoplankton are extremely diverse, varying from photosynthesizing bacteria (cyanobacteria), to plant-like diatoms, to armor-plated coccolithophores (drawings not to scale).Adapted

from Source:https://earthobservatory.nasa.gov/ContentFeature/Phytoplankton/images/phytoplankton_types.jpg

The evolution of marine photoautotrophs began in the Archaean period with the origin of photosynthesis. These primitive organisms are at the origin of the diverse photosynthetic biota from which all complex life is dependent. They are also at the origin of the oxygenation of the atmosphere and have profoundly modified the biochemistry of the oceans and the atmosphere (Simon et al., 2009).

DIVERSITY OF PHYTOPLANKTON

The term phytoplankton refers to the functional category of single-celled organisms (prokaryotes and eukaryotes) that perform oxygenic respiration. All marine phytoplanktonic prokaryotes are members of the Cyanobacteria. Cyanobacteria is phylum that belongs to domain Bacteria. The taxonomy of eukaryotic phytoplankton is extraordinarily diverse (Figure 2).

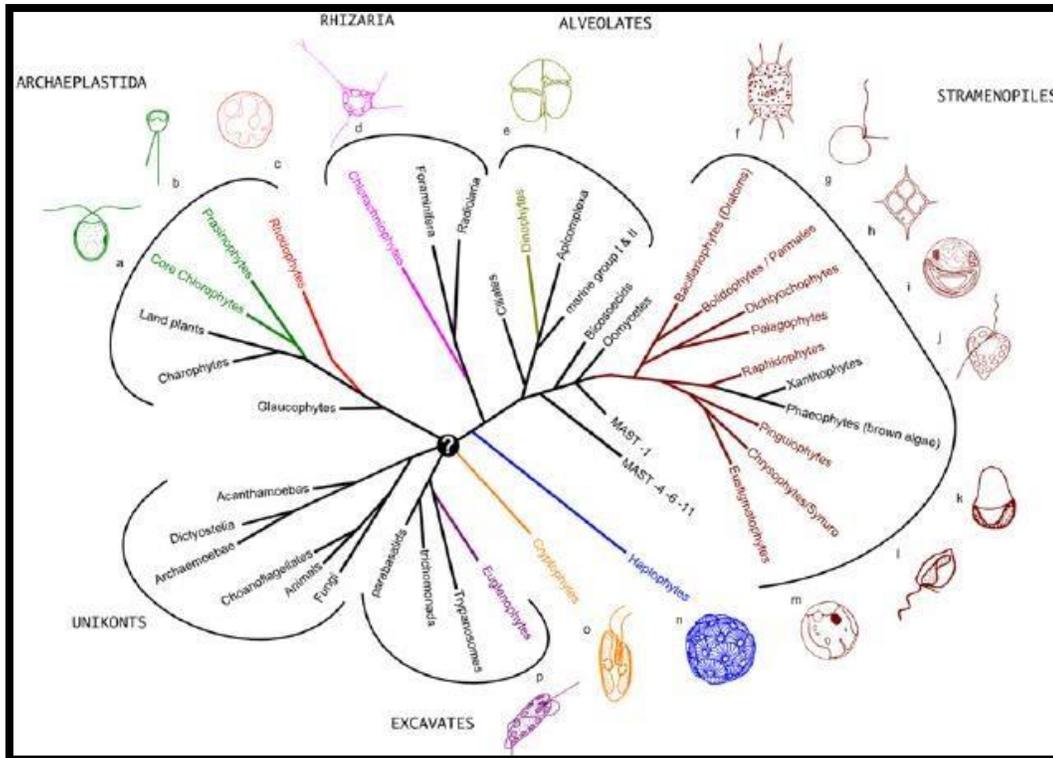


FIGURE 2. Schematic phylogenetic tree representing the distribution of phytoplanktonic taxa across eukaryote lineages (in color). Illustrations of (a) Chlorophyceae, (b) *Pseudoscurfieldia* sp., (c) *Porphyridium cruentum*, (d) *Gymnochloradimorpha*, (e) Dinoflagellates, (f) *Odontella* sp. (g) *Bolidomonas pacifica*, (h) *Dictyocha* sp., (i) *Aureococcus anophagefferens*, (j) *Heterosigma akashiwa*, (k) *Pinguiochrysis pyriformis*, (l) *Ochromonas* sp., (m) *Nannochloropsis salina*, (n) *Calcidiscus* sp., (o) *Cryptomonas* sp., (p) Euglenids (Adapted from Not et al., 2012).

The existing marine phytoplankton species can be found in the domains Bacteria and Eukarya of the tree of life. The diatoms, dinoflagellates, and haptophytes tend to predominate the phytoplankton communities on the continental shelves and are responsible for the seasonal blooms in temperate and polar waters among the currently recognised marine phytoplankton groups (Simon et al., 2009).

DIATOMS

Bacillariophyceae class of diatoms are ubiquitous phytoplanktons, unicellular and photosynthetic in nature. These microscopic, unicellular organisms have a distinctive cell structure known as frustule. Each frustule has two halves: an epitheca and a hypotheca. The diatoms include two orders: pennales (isobilateral symmetry) and centrales (radial symmetry). Their movements are phototactic and spontaneous type. They vary from 2 to 200 μm in size (Sharma et al., 2021). Silica cell walls (frustules), which are made up of two valves and separate girdle bands, are what give diatoms their distinctive appearance (Figure 3). In many moist terrestrial and aquatic environments, diatoms are the most common and diverse type of algae. Diatoms are vital components of marine and freshwater phytoplankton, benthos, and associated algal communities (Cameron, 2013).

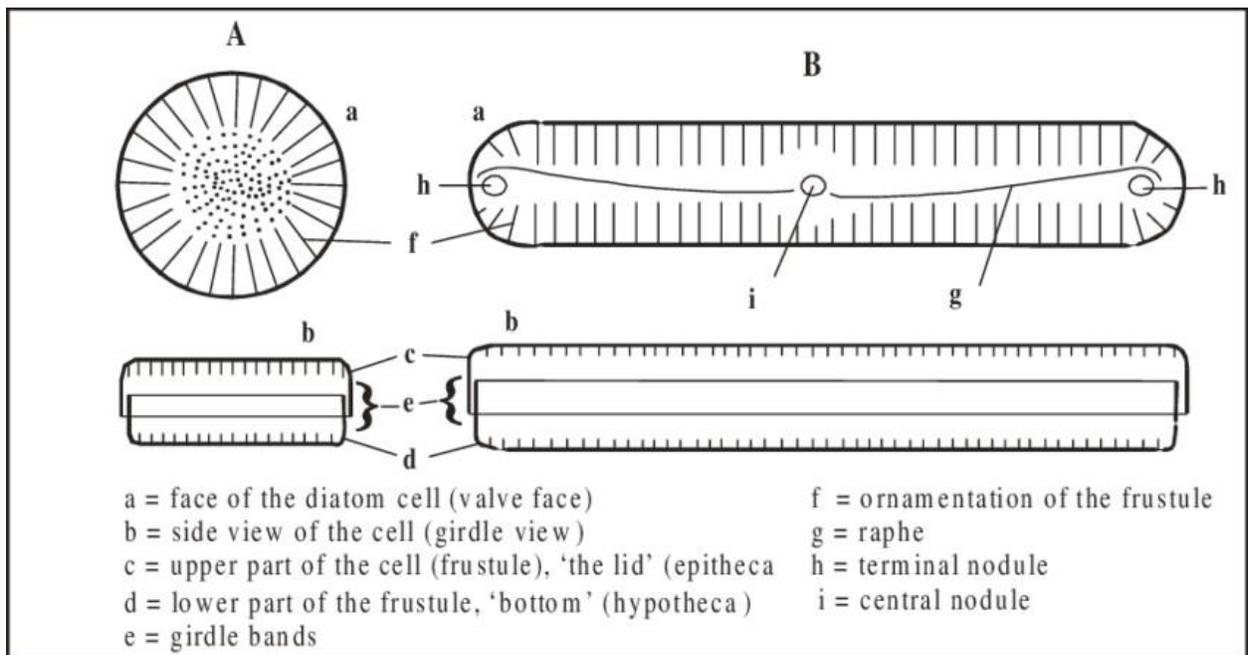


FIGURE 3. The structure of a diatom cell. Schematic representation of typical shapes of centric (A) and pennate (B) diatoms (Sorvari, 2001)

Diatoms can be planktonic, benthic, epiphytic, epizoid, endozoid, endophytic and can also live in air (Simon et al., 2009). Approximately 40% of all marine phytoplankton described species are diatoms, and they are extremely significant ecologically and biogeochemically, particularly in nutrient-rich systems. They often form colonies and chains. They are traditionally divided into two categories: Valve striae (rows of pores) are arranged basically in a central symmetrical pattern in centric diatoms, the symmetry can be unipolar or multipolar. Valve striae in pennate diatoms are essentially structured in relation to a line (Simon et al., 2009). The sequence of stages that each diatom species go through from its initial generation to its senescent cells makes up its life cycle (Sánchez et al., 2019) (Figure 4).

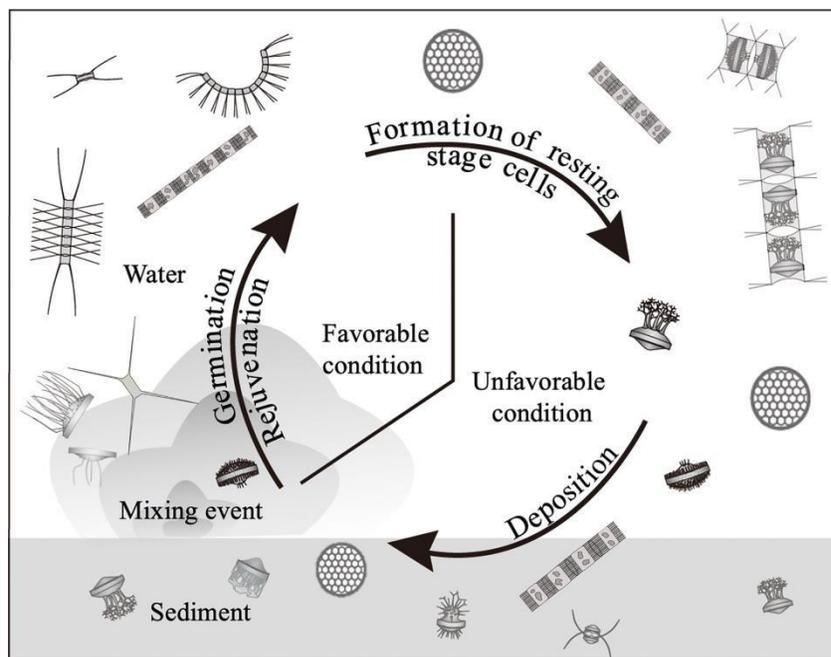


FIGURE 4. Conceptual diagram of life cycle of the centric diatoms forming the resting stage cells (resting spores and resting cells) (Adapted from Ishii et al., 2022).

DINOFLAGELLATES

In both fresh and salt water, the dinoflagellates (division Pyrrhophyta, class Dinophyceae) represent a significant group of phytoplankton. Dinophyta species can be free-living, planktonic, benthic, parasitic, or symbionts, however the majority of photosynthetic species are planktonic (**Simon et al., 2009**) (**Figure 5**). Their adaptation to a wide variety of environments is reflected by a tremendous diversity in form and nutrition and an extensive fossil record dating back several hundred million years. Toxins produced by some dinoflagellates can be harmful to humans, marine animals, fish, seabirds, and other members of the marine food web (**Van Dolah, 2000**). Some act as parasites or symbionts that depend on host species for a part of their nutrition; others are bioluminescent and emit light. An equal number of dinoflagellates obtain their carbon by ingesting other phytoplankton. Many are now being shown to be mixotrophic.

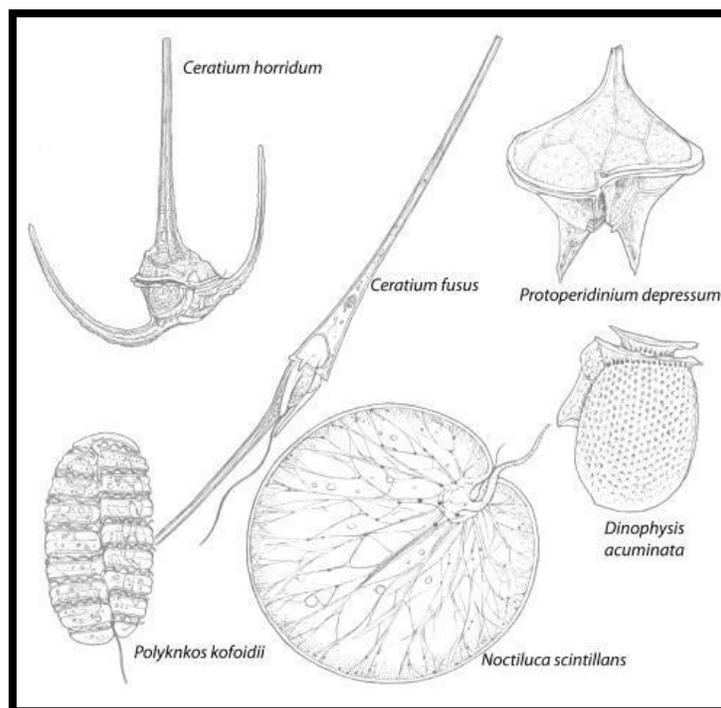


FIGURE 5. A variety of dinoflagellates from the NE Atlantic.

(Adapted from Dipper, 2016).

The main characteristic of dinoflagellates is the presence of a large nucleus, with permanently condensed chromosomes. The theca, which covers the cell wall, can be used to distinguish between two different cell types. A single layer of flattened vesicles is surrounded by an outer plasmalemma in the "naked" or unarmored forms. These cells are fragile and distort easily. The walls of the cells in armoured dinoflagellates are more rigid and inflexible due to the presence of cellulose or other polysaccharides inside each vesicle. These cellulose plates are arranged in distinct patterns (called "tabulation"), which are extensively used as taxonomic "fingerprints." Yet another distinguishing characteristic of dinoflagellates is that their motile cells have two unequal flagella. The cingulum, flattened ribbon-like flagellum which surrounds the cell in a transverse groove and provides propulsive and spinning force for the cell. The other flagellum, the sulcus, is directed posteriorly along a longitudinal groove and likely functions as a rudder for steering (Hackett et al., 2004; Simon et al., 2009) (Figure 6).

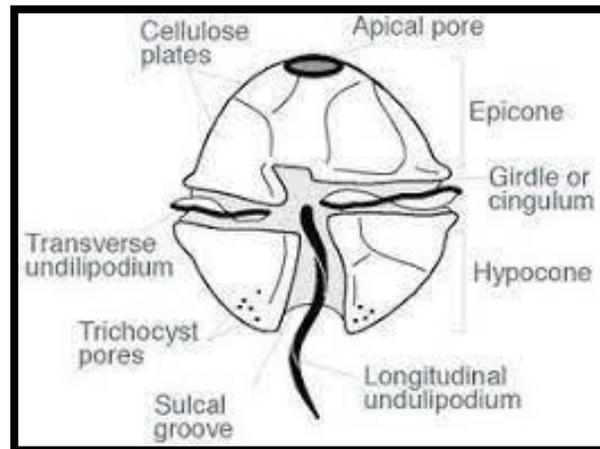


FIGURE 6. General structure of Dinoflagellate Adapted from

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Some dinoflagellates (called zooxanthellae) are capable of forming symbioses with a phylogenetically wide range of marine protists and invertebrate animals (Hackett et al., 2004). Dinoflagellates are responsible behind toxic algal blooms and are also significant marine primary producers and grazers. Many dinoflagellate species have been found to produce a range of natural toxins. Several of these toxins are effective at far lower concentrations than conventional chemical agents, although some of them can be exceedingly toxic (Wang, 2008). Many dinoflagellate species have been shown to produce potent neurotoxins, which are frequently associated to the phenomenon known as "red tides" (Haskettetal., 2004).

Dinoflagellates have an unusual genome consisting of liquid, crystal-like chromosomes; and a haplontic life cycle that often contains a non-motile, dormant cyst stage (Carty &Parrow, 2015) (Figure 7).

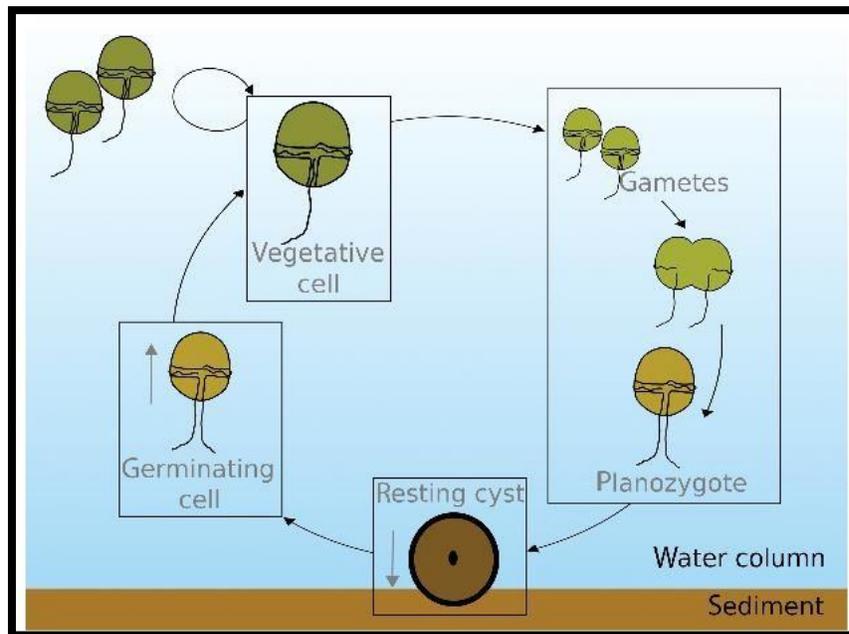


FIGURE 7. Conceptual overview of the dinoflagellate life cycle, the four model compartments (vegetative cells, gametes, cysts and germinating cells), and the transitions among the compartments(Adapted from Warns et al., 2012).

HAPTOPHYTES

The division Haptophyta is a group of unicellular algae that are predominantly marine, although there are a few freshwater and terrestrial records. Haptophyte nutrition is mainly phototrophic, but many exhibit phagotrophy and some are exclusively heterotrophic. Two classes make up the division Haptophyta: Pavlovophyceae, which have two unequal flagella and Prymnesiophyceae, which have two flagella that are more or less equal (Simon et al., 2009). The division Haptophyta is a group of unicellular algae that are predominantly marine, although there are a few freshwater and terrestrial records. A few haptophytes form colonies or short filaments, but the majority are solitary motile or nonmotile forms. In most cases, the cells are covered with scales of varying degrees of complexity, ranging from elaborate calcified formations termed as unmineralized organic scales, many of which can only be observed in electron microscopy (Eikrem et al., 2017).

Haptophyte algae account for 30-50% of total chlorophyll a biomass in modern oceans. By photosynthesis and calcification, calcifying haptophyte algae known as coccolithophores contribute significantly to the global carbon cycle by fixing dissolved inorganic carbon (DIC) (GranStadniczeňko et al., 2017; Tsuji & Yoshida, 2017). Haptophytes produce and accumulate a variety of compounds during photosynthesis, including acid polysaccharides (APs), mannitol, long-chain unsaturated ketones (alkenones), β -glucan, and dimethylsulphonylpropionate. Coccolithophores contribute in the limestone sequestration of atmospheric CO₂. Haptophytes exhibit a wide range of physical characteristics and carbon metabolism (Tsuji & Yoshida, 2017). The haptonema, a filiform organelle associated with the flagella sometimes employed for cell anchoring or prey capture, is the most recognisable characteristic of the majority of Haptophyta members. It is inserted between two smooth (nontinsel) flagella of equal or subequal length in most genera (Nicholls, 2003).

In particular, *E. huxleyi*, which is a highly abundant haptophyte in the ocean, has been considered a critical component of marine environments because of its dual capacity to fix environmental carbon via biomineralization (calcium carbonate, calcite) and through photosynthesis (Reyes-Prieto et al., 2009).

CHLOROPHYTA

Green algae are organisms that are characterized by having whiplash-shaped (smooth) flagella and chlorophyll a and b as the primary photosynthetic pigments, and starch located within the chloroplast as the primary storage product (Nozaki, 2003). Green algae, or chlorophyta, are classified according to the following characteristics: chloroplasts lacking an external endoplasmic reticulum; thylakoids typically arranged in stacks of two to six; chlorophyll-a and chlorophyll-b as photosynthetic pigments; true starch; and cellulosic walls or scales (Sheath & Wehr, 2003). The photosynthetic pigments are found in the thylakoids of the chloroplast. The cell wall of green algae is made up of an inner cellulose layer and a pectin layer.

CYANOBACTERIA

Cyanobacteria, popularly known as "blue-green algae," are photosynthetic prokaryotes that have been present on Earth for around 3500 million years (Mehdizadeh Allaf & Peerhossaini, 2022). They inhabit a variety of habitats, including freshwater, marine, brackishwater, hot springs, terrestrial ecosystems and symbiosis (with lichens, primitive animals and lichens). Cyanobacteria can survive and thrive in the most extreme environments, including geothermal environments, frozen systems, and hypersaline environments. Cyanobacteria can be unicellular, colonial, or multicellular filamentous. Their cell diameters range from less than 1 μm (Picocyanobacteria) to more than 100 μm (some tropical forms of the genus *Oscillatoria*) (Mehdizadeh Allaf & Peerhossaini, 2022).

Their total biomass has been calculated to be greater than 10^{15} g of wet biomass, with the majority of this mass being made up of the single-celled marine genera *Prochlorococcus* and *Synechococcus*, as well as the filamentous taxa *Trichodesmium* (a circumtropical marine form) and *Microcoleus vaginatus* and *Chroococcidiopsis* sp. Blooms of cyanobacteria are significant components of the environment and management of many nutrient rich fresh and brackish water basins. Certain cyanobacteria have the ability to fix nitrogen aerobically, making them significant participants in the biogeochemical nitrogen cycle of tropical oceans, terrestrial ecosystems, and some agricultural soils (**Garcia-Pichel, 2009**).

This group has members in the waters in both filamentous (heterocystous or not) and unicellular forms. In terms of both the carbon and nitrogen cycles, marine cyanobacteria are important for world ecology. Marine phytoplanktonic genera are dispersed within the Cyanobacterial tree. *Prochlorococcus* and *Synechococcus* are two closely related genera that are very prevalent and ubiquitous in the water, however they do not fix atmospheric nitrogen (**Simon et al., 2009**).

Cyanobacteria are essential primary producers and are found in aquatic environments. They may form biofilms and mats as well (benthic cyanobacteria). Cyanobacteria frequently form mass occurrences or "water blooms," in eutrophic water. Cyanobacteria were formerly called blue-green algae. Cyanobacteria in vast numbers can be toxic (**Sivonen, 2009**).

ECOLOGY

In the functioning of ecosystems and global ecology, phytoplankton plays a significant role. Almost half of the primary production on the earth is produced by phytoplankton species. Phytoplankton participates in the biological pump that contributes to the global carbon cycle by fixing carbon, part of which is then buried at depth (**Not et al., 2012**).

Moreover, phytoplankton form the foundation of almost all marine food webs. Certain phytoplankton taxa, particularly in coastal waters of temperate seas, have the ability to create large blooms under proper light and nutritional conditions. Harmful Algal Blooms, or HABs, are phytoplankton blooms that create toxins that damage higher trophic levels, having a major negative ecological and economic impact(Not et al.,2012).

Morphological examinations based on microscopy, flow cytometric cell counting, molecular surveys, and/or measures of the presence of certain substances produced by photosynthesis are used to study the ecology of phytoplankton communities. Diatoms, dinoflagellates, and coccolithophores are largely dominant in nutrient rich coastline and continental shelf waters, and these groups contain species with the ability of producing huge blooms. In addition to other biogeochemical effects, the ability of some phytoplankton in biomineralization of silica or calcium increases long-term carbon storage by increasing sinking to the sea floor during bloom occurrences. In addition to photosynthesis, mixotroph phytoplankton feed on prokaryotes or other tiny phytoplankton(Not et al.,2012).

OBJECTIVES

Considering the relevance of phytoplankton in the marine environment and their role in the ecosystem it would be interesting to study the diversity of phytoplankton in different areas.

Therefore the aim of the study is to analyze the different phytoplankton groups occurring in different areas of Goa.

The objectives are:

1. To study the diversity of phytoplankton in coastal waters of Goa.
2. To analyse the abundance of phytoplankton in coastal waters of Goa.

CHAPTER 2

MATERIALS AND METHODS

MATERIALS AND METHODS

2.1 Study site and sampling strategies

Sampling sites selected were Dona Paula, Miramar, Anjuna, Siridao, Caranzalem and ZuariMandovi estuary.

SR. NO.	LOCATION	COORDINATES
1.	Anjuna	15.5871° N, 73.7421° E
2.	Miramar	30.3744° N, 86.3586° W
3.	Dona Paula	15.4532° N, 73.8028° E
4.	Caranzalem	15.4665° N, 73.8048° E
5.	Siridao	15.4407° N, 73.8640° E
6.	Zuari estuary Mandovi estuary	15.4460°N, 73.82245°E 15.5047°N, 73.81161°E

Table 1: Location of sampling sites

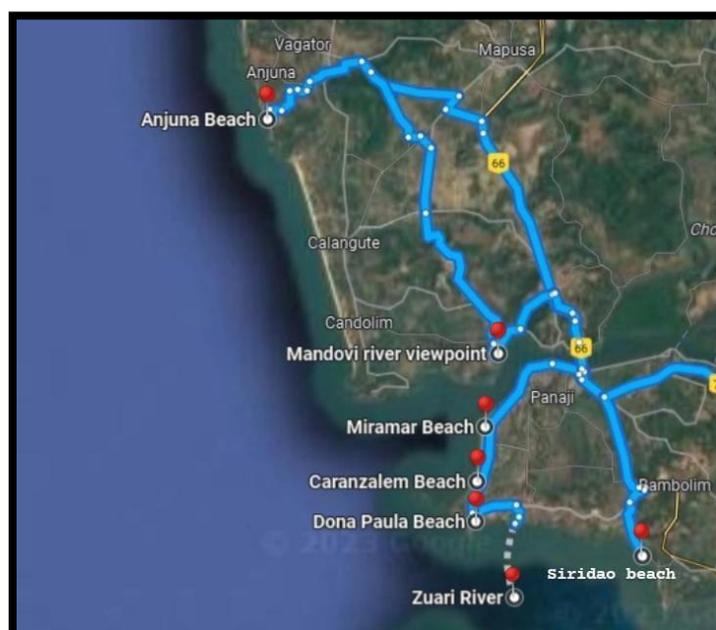


Figure 8. Geographic location of water collection sites

Water sample: The plastic bottles were rinsed with seawater before collecting the sample. Two litres of clearwater was collected in plastic bottles from each sampling stations. The bottles were filled by slowly submerging them into the water.

2.2 Physico-chemical analysis

The collected water samples were used for:

- 1) pH was recorded with the help of pH meter.
- 2) Salinity was recorded with the help of a refractometer.

2.3 Analysis of phytoplankton samples

1) Filtration

The water samples were filtered through 240 μ m mesh size sieve to exclude zooplanktons.

The same samples were then again filtered through 10 μ m mesh size. The mesh was then placed in a beaker and rinsed thoroughly with the filtrate itself. The content from the mesh was then transferred into a centrifuge tube.

2) Sample fixation

The Lugol's solution was prepared and stored in amber coloured bottle. The samples were fixed with this Lugol's solution and kept for settling in a centrifuge tube for 24 hours.



Figure 9. Lugol's solution used for fixation of sample

3) Siphoning of sample

With the help of an Intravenous tube the fixed samples were then siphoned upto the volume of 5 ml. The bottles were labeled properly and stored till further analysis.

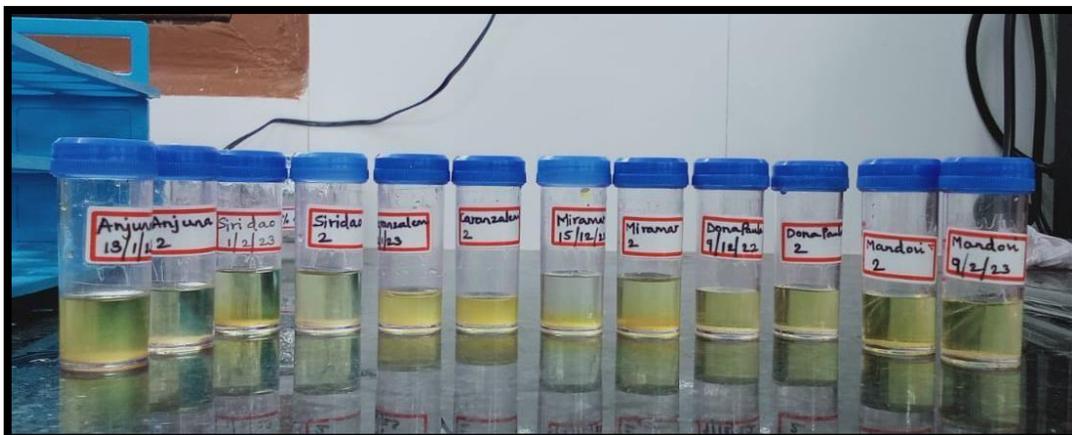


Figure 10. Siphoned samples

4) Microscopy

The samples were shaken well before examining. The siphoned samples were then transferred into Tarson's petriplate and were observed under 10X and 20X objectives of phase contrast microscope. The observed specimens were identified using phytoplankton identification keys (Perry,2010; [Kudela Lab](#) at the University of California Santa Cruz; Verlenkar,2004)

5) Counting method

The samples were shaken well before commencing the count. The cover slip was placed obliquely on the Sedgewick rafter. Approximately 1 ml of the sample was taken using a micropipette. The sample was then carefully put in the sedgewick rafter cell from one corner of the cell. The Sedgewick rafter cell was then left to stand on the microscope stage for 1520 minutes to allow the phytoplankton cells to settle. Fifty randomly selected grid squares/quadrants on the Sedgewick rafter cell were counted for each sample. The number of phytoplankton cells counted were recorded.



Figure 11. Sedgewick Rafter cell

The final calculation for total cell count in the sample was carried out using the formulae:

1) In 1 ml,

$$\text{Cell count} = (\text{DF}) (\text{NC})$$

Where, C – cells/ml

DF – Dilution factor

NC – Cells counted

2) In conc. sample (5 ml),

$$\text{Cell count} = \text{Total count in 1 ml} \times 5$$

3) In 1000 ml (1 L),

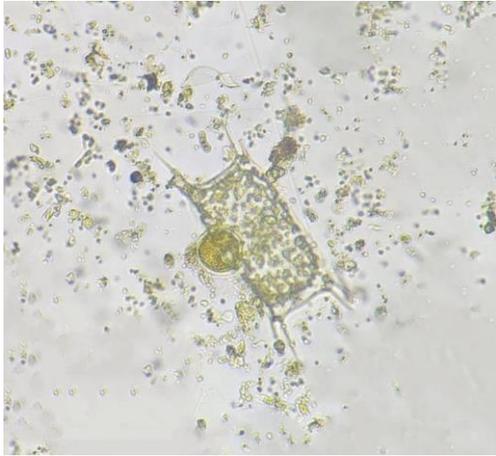
$$\text{Cell count} = \text{Cell count in 5 ml} \times 1000 \text{ ml} / 2000 \text{ ml}$$

CHAPTER 3

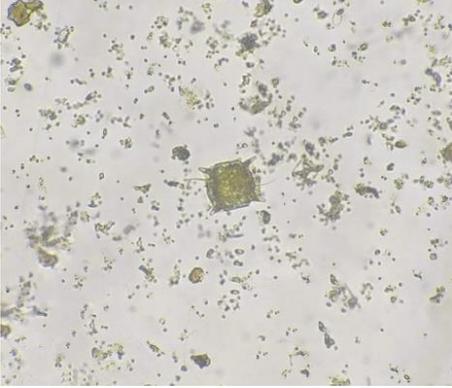
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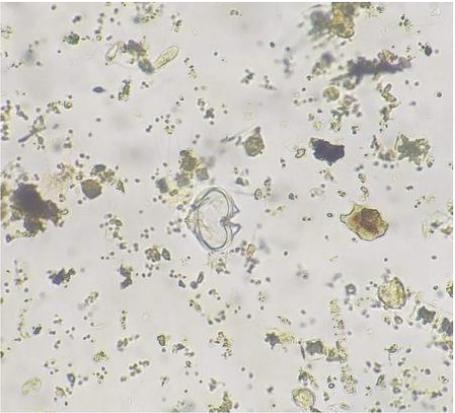
OBSERVATION

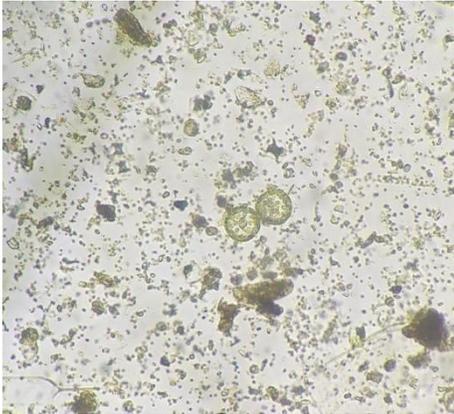
Table no. 2: Phytoplankton recorded in Anjuna sample

<p>Sample collected: 11/1/2023</p>		
SR.NO.	SPECIMEN	IDENTIFIED AS
1.		<p><i>Odontella sinensis</i></p> <p>Alternate name: <i>Biddulphia sinensis</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>
2.		<p><i>Coscinodiscus</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

<p>3.</p>		<p><i>Ceratium furca</i></p> <p>Alternate name: <i>Neoceratium furca</i></p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>
<p>4.</p>		<p><i>Navicula distans</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
<p>5.</p>		<p><i>Coscinodiscus</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

6.		<p><i>Coscinodiscus</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>
7.		<p><i>Peridiniumquinquecorne</i></p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>
8.		<p><i>Odontella</i> sp.</p> <p>Class: Biddulphiaceae</p> <p>Type: Centric diatom</p>

<p>9.</p>		<p><i>Odontella</i> sp.</p> <p>Class: Biddulphiaceae</p> <p>Type: Centric diatom</p>
<p>10.</p>		<p><i>Rhizosolenia</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>
<p>11.</p>		<p><i>Protoperidinium depressum</i></p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>

12.		<p><i>Detonula</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>
13.		<p><i>Pseudo-nitzschia</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
14.		<p><i>Cyclotella</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

15.

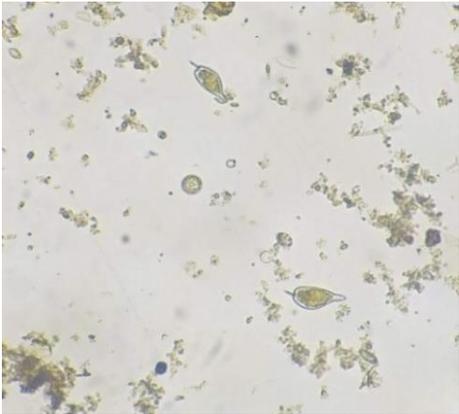


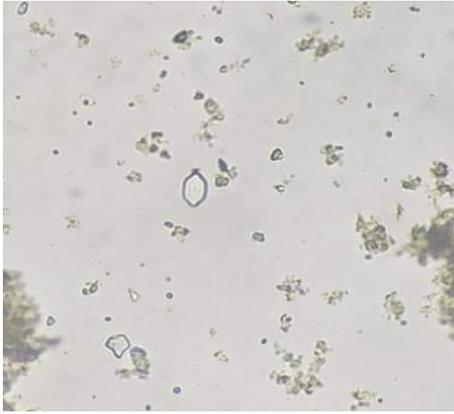
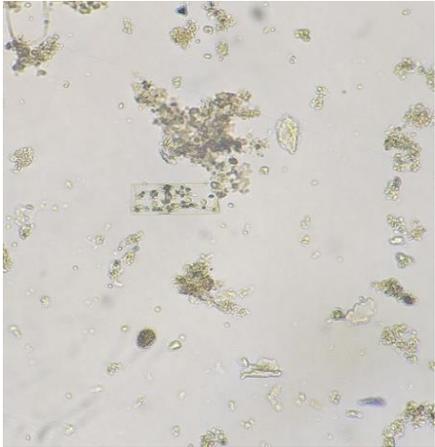
Thalassiosira sp.

Class: Bacillariophyceae

Type: Centric diatom

Table no. 3: Phytoplankton recorded in Caranzalem sample

<p>Sample collected: 20/1/2023</p>		
SR. NO.	SPECIMEN	IDENTIFIED AS
1.		<p><i>Prorocentrummicans</i></p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>
2.		Unidentified

3.		<p><i>Nitzschia panduriformis</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
4.		<p>Unidentified</p>
5.		<p><i>Ceratium tripos</i></p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>

6.	 <p>A light micrograph showing a chain of diatoms. The chain consists of several rectangular cells connected end-to-end. The cells have a distinct silica structure. The background is filled with other smaller, more irregular diatom fragments and organic matter.</p>	Unidentified
7.	 <p>A light micrograph showing a diatom with a characteristic naviculate shape. It is a single, elongated, spindle-shaped cell with a central constriction. The cell is surrounded by other diatom fragments and organic material.</p>	<p><i>Navicula</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>
8.	 <p>A light micrograph showing a silicoflagellate. The organism is a rectangular cell with a distinct silica structure. A black arrow points to the cell. The background contains other diatom fragments and organic matter.</p>	<p><i>Dictyochasp.</i></p> <p>Class: Dictyochophyceae</p> <p>Type: Silicoflagellate</p>

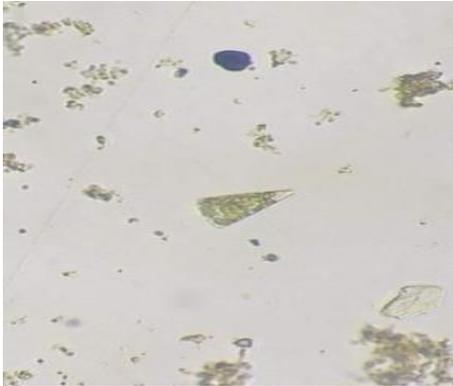
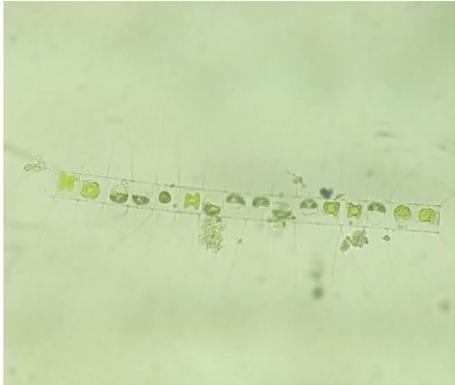
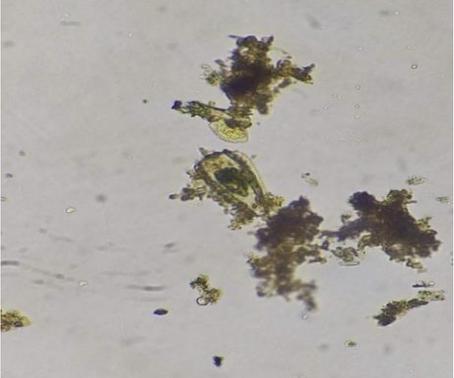
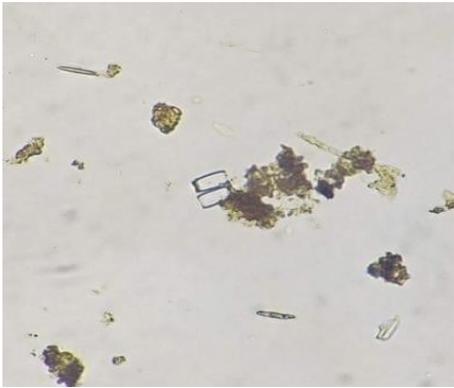
<p>9.</p>		<p><i>Protoperidinium</i> sp.</p> <p>Class: Dinophyceae</p> <p>Type: Dinoflagellate</p>
<p>10.</p>		<p>Unidentified</p>
<p>11.</p>		<p><i>Climacosphenia</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

Table no. 4: Phytoplankton recorded in Dona Paula sample

Sample collected: 9/12/2022		
SR. NO.	SPECIMEN	IDENTIFIED AS
1.		<i>Chaetoceros</i> sp. Class: Bacillariophyceae Type: Centric diatom
2.		Zooplankton group

3.		Unidentified diatom
4.		Unidentified
5.		Spine of a zooplankton

6.		Unidentified
7.		Tintinnid (zooplankton)
8.		Unidentified

9.	 <p>A light micrograph showing a curved chain of approximately 15-20 small, oval-shaped diatom cells. The cells are arranged in a slightly arcuate pattern. There are also some larger, more complex structures visible in the background.</p>	<p><i>Skeletonemas</i>p.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
10.	 <p>A light micrograph showing a central, elongated, spindle-shaped diatom cell. It is surrounded by several smaller, more irregularly shaped diatom fragments and clusters of cells.</p>	<p>Unidentified pennate diatom</p>
11.	 <p>A light micrograph showing a long, thin, needle-shaped diatom cell. It is surrounded by several larger, more complex diatom structures, including clusters and individual cells of various shapes.</p>	<p><i>Thalassiothrix</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>

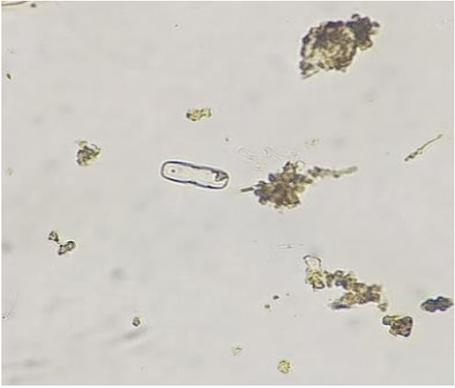
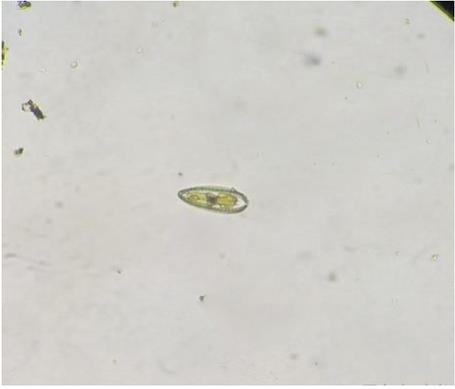
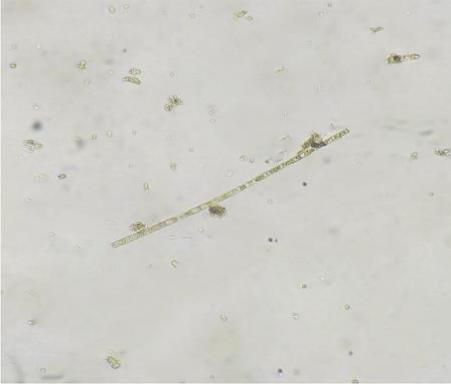
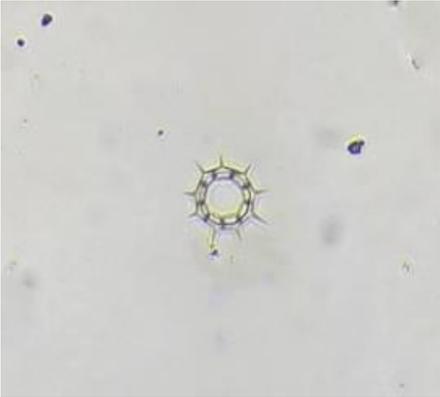
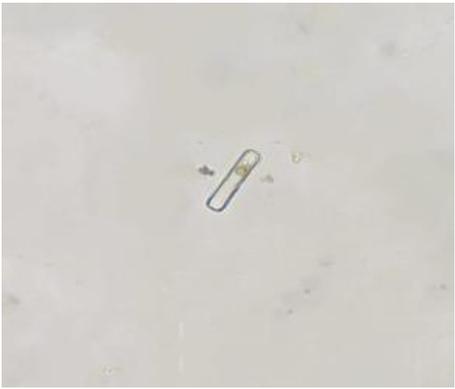
12.		Unidentified pennate diatom
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Table no. 5: Phytoplankton recorded in Miramar sample

Sample collected: 15/12/2022		
SR. NO.	SPECIMEN	IDENTIFIED AS
1.		<i>Surirella</i> sp. Class: Bacillariophyceae Type: Pennate diatom

2.		<p><i>Thalassiothrix</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
3.		<p><i>Navicula</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
4.		<p>Type: Silicoflagellate</p>

5.		<p><i>Planktoniella sol</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
6.		<p>Unidentified</p>
7.		<p><i>Chaetoceros</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

8.		Unidentified pennate diatom
9.		<i>Thalassionemanitzschioides</i> Class: Bacillariophyceae Type: Pennate diatom
10.		<i>Navicula</i> sp. Class: Bacillariophyceae Type: Pennate diatom

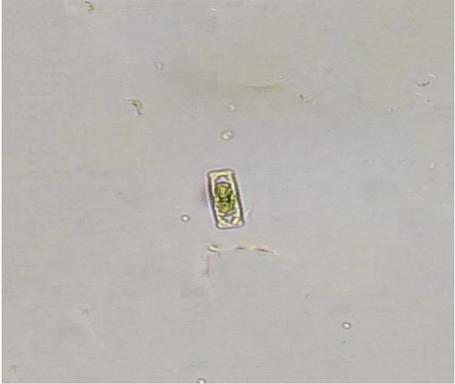
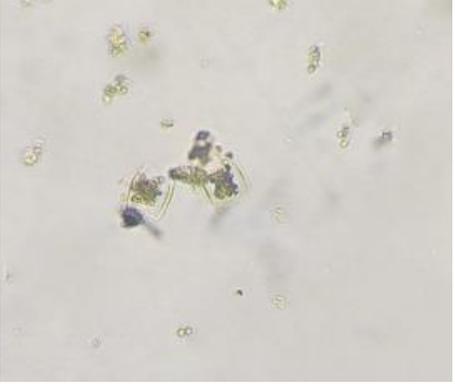
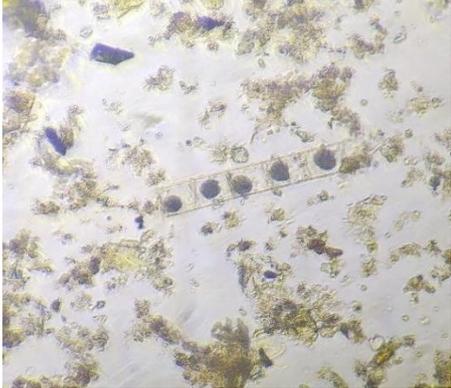
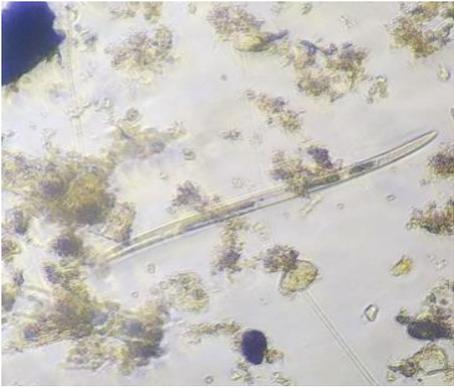
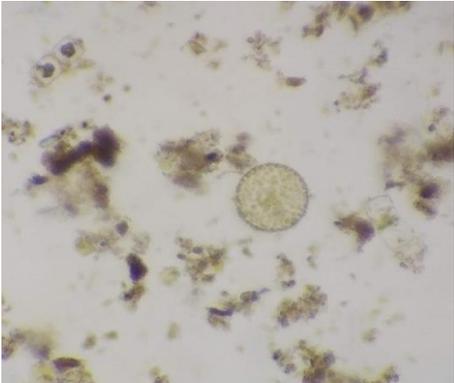
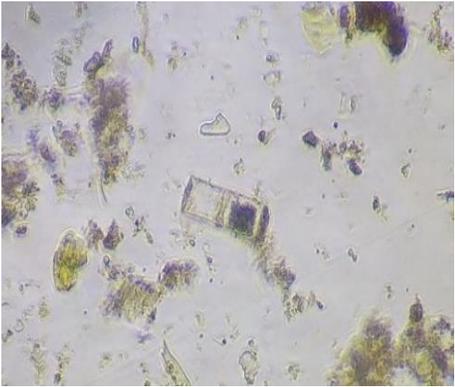
11.		Unidentified
12.		<i>Odontella</i> sp. Class: Bacillariophyceae Type: Centric diatom

Table no. 6: Phytoplankton recorded in Siridao sample

<p>Sample collected: 1/2/2023</p>		
SR. NO.	SPECIMEN	IDENTIFIED AS
1.		Unidentified diatom
2.		<p><i>Gyrosigma</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>

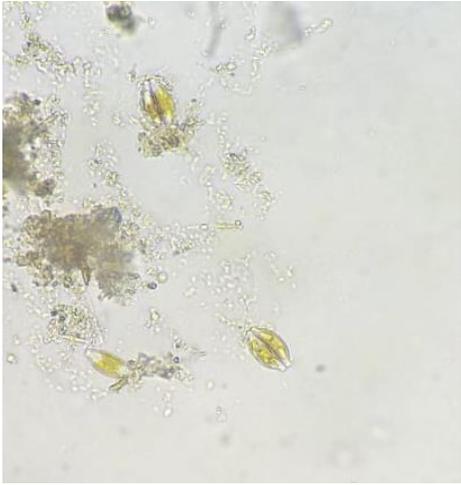
3.	 <p>A light micrograph showing several elongated, oval-shaped diatoms with a distinct central structure. The diatoms are stained, showing a yellowish-brown color with darker, almost black, central regions. They are scattered across a light-colored background.</p>	<p><i>Surirella ovate</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
4.	 <p>A light micrograph showing several circular diatoms. One prominent diatom in the center is clearly visible, showing a circular shape with a distinct central structure. The diatoms are stained, showing a yellowish-brown color with darker, almost black, central regions. They are scattered across a light-colored background.</p>	<p><i>Coscinodiscus</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

5.		Unidentified
6.		Unidentified

7.		Tintinnid (zooplankton)
8.		<i>Prorocentrum</i> sp. Class: Dinophyceae Type: Dinoflagellate

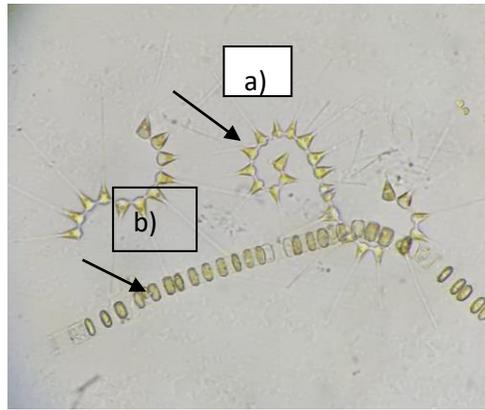
Table no. 7: Phytoplankton recorded in ZuariMandovi sample

Sample collected: 9/2/2023		
SR. NO.	SPECIMEN	IDENTIFIED AS

1.		Unidentified diatom
2.		<p><i>Amphora</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
3.		<p><i>Skeletonemacostatum</i></p> <p>Class: Bacillariophyceae</p> <p>Type: Centric diatom</p>

4.		<p><i>Navicula</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Pennate diatom</p>
5.		<p><i>Bacteriastrum</i> sp.</p> <p>Class: Bacillariophyceae</p> <p>Type: Centric Diatom</p>

6.



a) *Asterionellopsis glacialis*

Alternate name:

Asterionella japonica

Class: Bacillariophyceae

Type: Pennate Diatom

b) *Skeletonemacostatum*

Class: Bacillariophyceae

Type: Centric diatom

CHAPTER 4

RESULTS AND DISCUSSION

RESULTS

Salinity did not show any large variations and varied from 30-25 in all the six samples (Figure 12). pH varied from 7.2 – 8.09 in all the six samples (Figure 13).

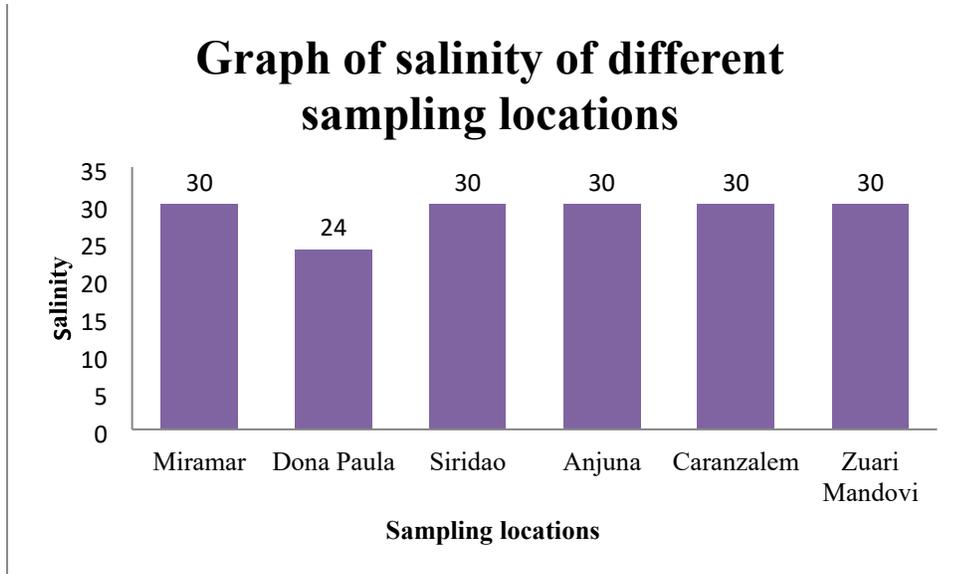


Figure 12. Salinity of water samples from all the sampling locations

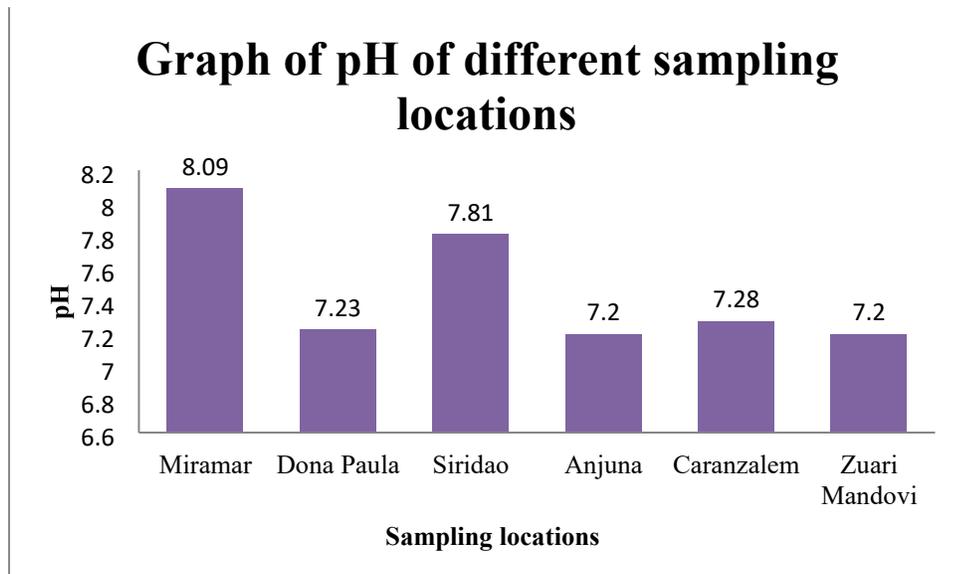


Figure 13. pH of water samples from all the sampling locations

Quantitative analysis of phytoplankton samples

The study focused on determining the abundance of phytoplankton in coastal waters of Goa.

Table 8: Abundance of phytoplankton cells/L

SR. NO.	SAMPLE	CELLS/ML
1.	Miramar	7,000 cells/L
2.	Siridao	17,750 cells/L
3.	Anjuna	21,600 cells/L
4.	Caranzalem	17,200 cells/L
5.	Dona Paula	5,250 cells/L
6.	ZuariMandovi	3,950 cells/L

Phytoplankton abundance showed significant difference between all the six samples. The highest number of phytoplankton were found in Anjuna sample with 21,600 cells/L followed by Siridao sample with 17,750 cells/L. The least number of phytoplankton were found in ZuariMandovi sample with 3,950 cells/L.

Diversity of phytoplankton

Most of the phytoplankton species observed from the samples belonged to diatoms and dinoflagellate groups. A total of 26 phytoplankton species were identified from all the six samples out of which 20 belonged to Diatom group, 4 belonged to Dinoflagellate group and 2 belonged to Silicoflagellate group. The observed phytoplankton belonged to classes Bacillariophyceae, Dinophyceae, Biddulphiaceae and Dictyochophyceae. The common

species found in the samples were *Coscinodiscus* sp. and *Navicula* sp. belonging to class Bacillariophyceae. Dinoflagellates were not observed in Miramar and Mandovi Zuari samples.

Table 9: Species observed in samples

SR. NO.	SPECIES	SAMPLE					
		Anjuna	Caranzalem	Siridao	Dona Paula	Miramar	ZuariMandovi
1.	<i>Amphora</i> sp.						
2.	<i>Asterionellopsis</i> sp.						
3.	<i>Bacteriastrum</i> sp.						
4.	<i>Coscinodiscus</i> sp.						
5.	<i>Cyclotella</i> sp.						
6.	<i>Climacosphenia</i> sp.						
7.	<i>Chaetoceros</i> sp.						

8.	<i>Ceratium</i> sp.						
9.	<i>Detonula</i> sp.						
10.	<i>Gyrosigma</i> sp.						
11.	<i>Navicula</i> sp.						
12.	<i>Nitzschia</i> sp.						
13.	<i>Odontella</i> sp.						
14.	<i>Pseudo-nitzschia</i> sp.						
15.	<i>Planktoniella</i> sp.						
16.	<i>Protoperidinium</i> sp.						
17.	<i>Prorocentrum</i> sp.						
18.	<i>Peridinium</i> sp.						
19.	<i>Rhizosolenia</i> sp.						
20.	<i>Surirella</i> sp.						
21.	<i>Skeletonema</i> sp.						
22.	<i>Thalassionema</i> sp.						
23.	<i>Thalassiothrix</i> sp.						
24.	<i>Thalassiosira</i> sp.						

DISCUSSION

The number of phytoplankton is influenced by environmental conditions such as pH, salinity, nutrient availability, the type of water, etc. (Sun et al., 2022). The samples collected from different stations were used for determining the pH, salinity and checking the diversity and the abundance of phytoplankton. The result showed that the pH of the water samples was between 7-8.5 which has neutral to alkaline properties. According to Haroon et al. (2017) pH levels between 8.0 to 8.6 indicate exceptional alkaline conditions for phytoplankton to flourish in water. Reduced pH values may limit phytoplankton primary production (Berge et al., 2010). Salinity plays a key role in regulating the metabolism and reproductivity of organisms. (Kinne, 1971). Salinity value was found to be within the normal range.

After filtration the samples were stored by fixing them with Lugol's solution. For long term analysis phytoplankton sample should be preserved in fixatives and preservatives. Lugol's solution is an excellent preservative, particularly for retaining the flagella and cilia of flagellated and ciliated phytoplankton, as well as increasing their density and causing them to sink to the bottom. Diatoms and dinoflagellates are two of the most important phytoplankton types in the ocean (Bi et al., 2021). The present study revealed enormous diversity in the diatoms and dinoflagellates inhabiting the coastal waters of Goa. The total number of phytoplankton listed in each of the six sampling locations varied considerably. Pratiwi et al. (2017) showed that the phytoplankton composition based on number of species in the waters of the Bali Strait represent three classes, i.e. Bacillariophyceae, Dinophyceae, and Cyanophyceae. In the present study, the species observed belonged to classes Bacillariophyceae, Dinophyceae, Dictyochophyceae and Biddulphiaceae with Bacillariophyceae found to be the most diverse class in the coastal waters of Goa.

Changes in nutrient concentrations had a considerable impact on diatom-dinoflagellate competition, resulting in diatom competing the dinoflagellates at high nutritional concentrations (Bi et al., 2021). This suggests that in present study the locations where dinoflagellates were not found, the nutrient concentration might be the reason for diatoms to outcompete the dinoflagellates.

Water quality has a significant impact on the quantity, communities, and biodiversity of phytoplanktonic cells (Alprol et al., 2021). A number of 1 cell/L indicates poor water quality, a value between 1 and 40 cell/L indicates intermediate water quality, and a value greater than 40 cell/L indicates high water quality (Cahyonugroho et al., 2022). The present study showed abundance of phytoplankton in a range of 3,000-25,000 cells/L of all the six samples suggesting that the water quality is high of all the six locations.

SUMMARY

Phytoplankton are known to respond to environmental factors and influence the environment, thereby can be used to determine the quality of water. Therefore, it is necessary to understand phytoplankton composition in order to measure the production patterns in marine ecosystems. The phytoplankton diversity of Anjuna, Siridao, Miramar, Caranzalem, Dona Paula and ZuariMandovi estuary was studied. The samples were collected in 2 L plastic bottles and the further analysis were carried out in the laboratory. pH and salinity were determined using pH meter and refractometer respectively. Salinity in all the six samples showed variation from 30-25 while pH varied from 7.2-8.09. After filtering through 240 μm and 10 μm mesh size sieves the water was fixed with Lugol's solution and the phytoplankton were observed under the microscope and the cells were counted using a Sedgewick rafter cell. Different species were observed under the microscope belonging to classes Bacillariophyceae, Dinophyceae, Dictyochophyceae and Biddulphiaceae. Phytoplankton

cells were counted and calculated (cells/L) that showed Anjuna sample to be the richest in phytoplankton density followed by Siridao sample. Dona Paula and ZuariMandovi samples showed the least number of phytoplankton cells. All the samples showed large diversity containing different species of diatoms and dinoflagellates. As phytoplankton respond quickly to environmental they can be used as biological indicators by studying diversity and taxonomy. Moreover, they form the base of the marine food web and therefore are essential to maintain the balance in marine ecosystem.

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