2.1 Description of the study area

Vembanad Kol Wetland was included in the list of wetlands of international importance, as defined by the Ramsar Convention for the conservation and sustainable utilization of wetlands. It is a home to more than 20,000 waterfowls and serves as an ideal habitat for shrimps. Major livelihood activities of the people living on the shores of the lake include: agriculture, fishing, tourism, inland navigation, coir retting and lime shell collection. Uncontrolled mining of shells from the lakebed is also posing a threat to the ecosystem. Vembanad Lake extends from Alappuzha in the south to Azheekode in the north. This wetland system covers an area of over 2033.02 km². The lake is fed by 10 rivers flowing into it including the six major rivers of central Kerala namely: Achenkovil, Manimala,
Meenachil, Muvattupuzha, Pamba and Periyar. There are three islands located in the lake viz., Pathiramanal, Perumbalam and Pallippuram. River Periyar evacuates into the Cochin backwater system in the north and Muvattupuzha River in the central part.

Cochin backwater system situated at the tip of the northern Vembanad lake is regarded as a positive tropical estuary located between 9° 14’ to 10° 12’N and 76° 10 to 76° 36’E with its northern boundary at Azheekode and southern boundary at Thannermukkom bund. The estuary has a length of 80 km and width varies from 500 to 4000m. It has been regarded as the second largest estuarine system in India, fed by six rivers with fresh water discharge of about 291,010 m³ per year (Srinivas et al., 2003). It is permanently connected to Arabian Sea by 450 m wide channel at the bar mouth. There are three seasonal conditions prevailing in the estuary viz., pre monsoon (PRE), monsoon (MON) and post monsoon (POM). During premonsoon season (February-May), warm climate prevails over the coast, runoff is least and the estuary is predominantly marine in nature. The environment is more or less stable, well mixed and homogenous water mass is present. This estuary is under the profound influence of monsoon (June - September). It has been reported that about 71% of annual rainfall is contributed by monsoon (Jayaparakash, 2002). Hydrobiological studies (Menon et al., 2000) revealed that the high flushing during monsoon completely transforms the estuary into a freshwater habitat. Postmonsoon (October-January) is generally the stabilization period and is characterized by diminished river discharge and tides gradually gain momentum as the estuarine condition change to partially mixed type.
The rapid industrialization and the increase in population around the Cochin estuary have resulted in the discharge of a heavy load of the inorganic and organic wastes. The pollutants of greatest concern in the estuarine ecosystem are those which are persistent, such as toxic heavy metals, insecticides and pesticides. Organic materials such as domestic sewage and food processing wastes are rapidly decomposed and often enrich the elements essential for plant nutrition and productivity. Pollutants from industrial units like the Fertilizers and Chemicals Travancore Limited (FACT), Indian Rare Earths (IRE), Hindusthan News Print Factory, Hindusthan Organic Chemicals (HOCL), BPCL-Kochi refinery and Cochin Port area (handling large quantities of sulphur, crude oil and other petroleum products) find their way to this backwater system. Domestic sewage channels, coconut husk retting yards also contribute huge loads of organic matter and nutrients into the water body every year (Stephen, 1985).

2.2 Sampling and analytical methodology

Seasonal samplings of water, sediments and fish landings were carried out from the fifteen selected stations (S1 to S15) (figure 2.1) spread across Cochin backwater system. The samples were collected in five sampling campaigns that were scheduled during January 2009, April 2009, August 2009, January 2010 and April 2010, representing three seasons (premonsoon, monsoon and postmonsoon). Water samples (both surface and bottom layers) were collected using Niskin Sampler and surface sediment samples were taken using Van Veen Grab. Water samples for general hydrography and nutrient analysis was subsampled into high density polyethylene bottles, kept on ice bags transported to laboratory and analysed without delay.
Figure 2.1 Location of sampling stations
Table 2.1 General features of sampling stations

<table>
<thead>
<tr>
<th>Station Code</th>
<th>Place</th>
<th>Average Depth (m)</th>
<th>Latitude</th>
<th>Longitude</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>S1</td>
<td>Karippadam</td>
<td>4.3</td>
<td>9°47.646’N</td>
<td>76°25.708’E</td>
<td>Thickly populated area with outflow of domestic wastes</td>
</tr>
<tr>
<td>S2</td>
<td>Murinjapuzha Enadi</td>
<td>4.4</td>
<td>9° 47.887’N</td>
<td>76° 24.607’E</td>
<td>Disposal of domestic wastes</td>
</tr>
<tr>
<td>S3</td>
<td>Murinjapuzha Brahmmamangalam</td>
<td>4.9</td>
<td>9° 48.495’N</td>
<td>76° 24.019’E</td>
<td>Disposal of domestic wastes</td>
</tr>
<tr>
<td>S4</td>
<td>Murinjapuzha</td>
<td>4.6</td>
<td>9° 49.508’N</td>
<td>76° 23.359’E</td>
<td>Disposal of domestic wastes</td>
</tr>
<tr>
<td>S5</td>
<td>Perumbalam</td>
<td>3.2</td>
<td>9° 49.793’N</td>
<td>76° 21.430’E</td>
<td>Disposal of Domestic and fish processing wastes</td>
</tr>
<tr>
<td>S6</td>
<td>Aroor-Kumbalam</td>
<td>5.6</td>
<td>9° 52.105’N</td>
<td>76° 18.409’E</td>
<td>Fishing and processing unit operations</td>
</tr>
<tr>
<td>S7</td>
<td>Thevara Bridge</td>
<td>2.7</td>
<td>9° 55.070’N</td>
<td>76° 18.253’E</td>
<td>Sewage Outfall</td>
</tr>
<tr>
<td>S8</td>
<td>Marine Science Jetty</td>
<td>3.3</td>
<td>9° 57.77’N</td>
<td>76° 16.919’E</td>
<td>Sulphur Jetty input and sewage, Industrial pollution</td>
</tr>
<tr>
<td>S9</td>
<td>Bolgatty</td>
<td>2.5</td>
<td>9° 59.213’N</td>
<td>76° 16.064’E</td>
<td>Inland navigation and other tourism operations, waste disposal</td>
</tr>
<tr>
<td>S10</td>
<td>Mulavukad</td>
<td>2.2</td>
<td>10° 01.857’N</td>
<td>76° 15.789’E</td>
<td>Disposal of domestic sewages and fish wastes</td>
</tr>
<tr>
<td>S11</td>
<td>Chenoor</td>
<td>1.9</td>
<td>10° 03.255’N</td>
<td>76° 16.043’E</td>
<td>Domestic sewages outfall</td>
</tr>
<tr>
<td>S12</td>
<td>Cheranelloor</td>
<td>2.1</td>
<td>10° 03.999’N</td>
<td>76° 16.924’E</td>
<td>Disposal of domestic sewage and wastes</td>
</tr>
<tr>
<td>S13</td>
<td>Eloor</td>
<td>4.4</td>
<td>10° 05.656’N</td>
<td>76° 17.049’E</td>
<td>Industrial Belt</td>
</tr>
<tr>
<td>S14</td>
<td>Edayar</td>
<td>2.5</td>
<td>10° 05.502’N</td>
<td>76° 17.744’E</td>
<td>Industrial Belt</td>
</tr>
<tr>
<td>S15</td>
<td>FACT-Kalamassery</td>
<td>2.8</td>
<td>10° 04.993’N</td>
<td>76° 17.906’E</td>
<td>Industrial Belt</td>
</tr>
</tbody>
</table>

2.2.1 General hydrographic parameters

Hydrographic parameters such as pH, dissolved oxygen, carbon dioxide, temperature, salinity, alkalinity, hardness, rainfall, depth and transparency were determined. Nutrients like nitrite-N, nitrate-N, ammonia-N, phosphate, silicate, iron were also estimated in the water samples.
Rainfall data were obtained from the meteorological Centre, Trivandrum (Govt. of India). Tide data was collected from Cochin Port Trust (Govt. of India). Measurement of transparency of water column was carried out with Secchi disc and extinction coefficient was calculated using the formula (Michael, 1984).

Extinction Coefficient = \( \frac{1.7}{\text{depth in meters}} \)

pH measurements of water samples were made using a portable pH meter (Perkin Elmer, accuracy, ± 0.01). Temperature was measured using a precision mercury thermometer graduated from 0-50°C with accuracy of ±0.01°C. Immediately after collecting the water sample in a narrow mouthed polyethylene bottle, the thermometer was introduced into the water column upto 5 cm. Salinity was estimated by Mohr-Knudsen method (Muller, 1999). Modified Winkler method was used for the estimation of dissolved oxygen (Hansen, 1999). Free CO\(_2\) was determined with NaOH reagent and phenolphthalein indicator (Golterman et al., 1978). Alkalinity of the water samples were estimated by the method of Koroleff (Anderson et al., 1999). Five day biochemical oxygen demand test (BOD\(_5\)) employed for determination of BOD values of water samples (APHA, 1995). The dissolved oxygen in sample was determined before and after 5 days of incubation.

### 2.2.2 Estimation of nutrients in water column.

Nutrients (nitrite, nitrate, phosphate and silicate) were estimated spectrophotometrically using standard methods.

**Nitrite**

\( \text{NO}_2^- \) was estimated using the standard procedure suggested by Grasshoff et al., (1999) in which the nitrite formed an azodye with
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sulphanilamide and N-1 naphthyl ethylene diamine dihydrochloride. The spectrophotometric determination was done using UV-Vis spectrophotometer (Genesys 10 UV- Thermospectronic).

Nitrate

$\text{NO}_3^- \text{ was reduced to NO}_2^- \text{ using copper coated cadmium granules and determined as nitrite as outlined above (Grasshoff et al., 1999).}$

Dissolved inorganic phosphate

All methods for the determination of dissolved inorganic phosphate in water samples is based on the reaction of the ions with an acidified molybdate reagent to yield a phosphomolybdate heteropoly acid, which is then reduced to a intense blue coloured compound (Grasshoff et al., 1999). A known volume of sample was treated with mixed reagent and acidified ascorbic acid and the absorbance of the resulting blue complex was measured at 880nm using spectrophotometer.

Silicate

The determination of dissolved silicon compounds in natural waters is based on the formation of a yellow silicomolybdic acid when an acid sample is treated with a molybdate solution. This complex was reduced with ascorbic acid and absorbance was measured at 880nm (Grasshoff et al., 1999).

Iron

Water samples were preconcentrated according to Grasshoff et al., (1999); concentration of iron was estimated by Atomic absorption spectrometer (Perkin Elmer 3110).

2.2.3 General sediment characteristics
Each sample were transferred to polythene bottles using a plastic spatula and made airtight and taken to the laboratory for the further analysis under ice-cold storage conditions. Approximately, 200 g of sediment from each sample was dried in Freeze drier equipment (Beetta instruments and equipments, Chennai). Dried sediments were homogenized using a mortar and pestle, sieved through a 230 mesh sieve and used for the analysis of total organic carbon, CHNS analysis, nutrients and biochemical composition.

pH of the fresh wet sediments was measured in situ using a potable pH meter calibrated with buffer solutions. Redox potential of the fresh wet sediment was measured in situ using a portable Eh meter employing Zobell’s solution for the calibration of the electrodes (Brassard, 1997).

Texture analysis of sediments

The textural characteristics of the sediments were determined by pipette analysis (Folk 1980), after removing the inorganic carbonates using 10% HCl and organic matter using H2O2. Sediment was wet sieved through a 63-µm sieve to collect the sand fraction. The mud fraction was divided into silt (63-4 µm) and clay (<4 µm) fractions by timed gravimetric extraction of the dispersed sediments.

Total Organic Carbon

Total organic carbon (TOC) was also determined using a CHN analyzer after removing the inorganic carbon with 1 N HCl (Tung and Tanner, 2003).

Total Organic Matter (TOM)

TOM was obtained by multiplying the Total organic carbon values with 1.724 (Nelson et al., 1996).
2.2.4 Fractionation of nitrogen in sediments

The analyses of nitrite, nitrate and ammonia were carried out by the “KCl equilibrium extraction method” (Agemian, 1997) which involves shaking of the sediment sample with a solution of KCl (2N) at room temperature for a period of one hour followed by filtration using standard filtration equipment and Whatman 42 filter paper. The filtrate containing nitrogen in the dissolved state was stored at 4°C until analysis. From the filtrate the nitrite, nitrate, ammonia and were quantitatively estimated by spectrophotometric analyses as described earlier. Total nitrogen in sediments was determined by the following procedure. Sediment samples were digested in sulphuric acid in the presence of potassium sulphate and copper sulphate catalyst (Agemian, 1997). Organic compounds of nitrogen as well as free inorganic forms were thus converted to ammonium ions which were determined spectrophotometrically using indophenol blue method. Total nitrogen was estimated by CHNS analyser (Vario EL III CHNS Analyzer.).

2.2.5 Fractionation of phosphorous in sediments

The sequential extraction scheme by Golterman, (1996) using chelating agents was employed for estimating different phosphorus fractions (figure 2.2). Compared with the other methods, chelating agents allow a specific extraction of inorganic phosphorus with less destruction of organic phosphorus (Golterman, 1996). Iron bound phosphorus (Fe-IP) was extracted with buffered Ca-EDTA/dithionite and calcium bound fraction (Ca-IP) subsequently with Na-EDTA. In the next step, acid soluble organic phosphorus (ASOP) was extracted with H₂SO₄ and then alkali soluble organic
phosphorus (Alk-OP) with 2M NaOH at 90°C for 2 hours. Residual organic phosphorous (ROP) was measured after 1 hour K₂S₂O₈ digestion in acid medium. All the extractions were carried out under mild continuous shaking and the results are expressed on the dry weight basis. Generally, iron and calcium bound inorganic fractions and acid soluble organic fractions of phosphorous are considered to be bioavailable (Diaz-Espejo et al., 1999).

Figure 2.2 Sequential extraction scheme for phosphorus fractionation

2.2.6 Biochemical composition of sediments

Spectrophotometric methods were employed for the determination of biochemical compounds in sediments. Proteins (PRT) analyses were carried out following the procedure of Lowry et al., (1951), as modified by Rice, (1982) to account for the reactivity of phenolic compounds, with albumin as the standard. The amount of protein nitrogen was obtained by
multiplying protein with a factor of 0.16 (Mayer et al., 1986). Total carbohydrates (CHO) were analyzed according to Dubois et al., (1956), using glucose as the standard. Total lipids (LPD) were extracted according to Bligh and Dyer (1959), and estimated according to Barnes and Blackstock, (1973) using Cholesterol as the standard. All the analyses were carried out in triplicates and the average value was reported. The sum of all PRT, CHO and LPD was defined as the labile or easily assimilable organic fraction (Danovaro et al., 1993; Cividanes et al., 2002). Tannin and lignin in sediments were extracted using 0.05M NaOH at 60° for 90 minutes and the estimated spectrophotometrically by the sodium tungstate-phosphomolybdic acid method (Nair et al., 1989; APHA, 1995), using tannic acid as the standard. The principle involved is the development of a blue colour on reduction of Folin phenol reagent by the aromatic hydroxyl groups present in tannins and lignins. The effects of Mg and Ca hydroxides and/or bicarbonates present in the seawater were suppressed by the addition of trisodium citrate solution (Nair et al., 1989). Chlorophyll pigments in sediments were determined spectrophotometrically after extracting with 90% acetone according to (Lorenzen, 1967; APHA, 1995).

2.2.7 Primary Production

Plankton sampling and Enumeration

The procedure adopted in the present study was phytoplankton count based on Phytoplankton Identification Manual [Fritsch, (1935); Prescott, (1969); Santhanam et al., (1975); Verlencar and Somshekar Desai, (2004); Mitra et al., (2006); Dona et al., (2004)]. Photographs of the identified phytoplanktons in the study area are shown in figure 2.3. Phytoplankton production and distribution were examined over a period of 15 months for phytoplankton ecology from 15 sampling stations. In order
to prevent the decay of the samples the fixation and preservation was carried out. The preserved samples are immediately stored in the high quality polyethylene bottles and neatly labeled. The labeling specifies the date, time, location of the station and type of gear used. Record was kept for cells in good or vigorous condition and those in the poor condition.

Water samples were collected using Niskin sampler and 50L of water was filtered through phytoplankton net of 20µm mesh size made of bolting silk. The filtrate was preserved in 4% neutralized formaldehyde/Lugol’s iodine solution. The buffered formalin was prepared by adding 30gm Borax b (Sodium tetraborate) into 1L formalin. The pH of the preserving medium was maintained at about 8.2. The preservation was done onboard immediately after collection. Quantitative analysis was done employing Sedgewick-Rafter cell. A settling and siphoning procedure was followed to concentrate samples from 250 ml to 10 ml (Utermohl, 1958). Species identification was done using a Nikon E200 light microscope. For counting phytoplankton cells and identification of genera and species, two 1ml replicates of concentrated samples were transferred into Sedgewick-rafter cell and examined microscopically at 200x magnification; the whole slide (1000 fields) was counted for diatoms, dinoflagellates and silicoflagellates.

For analyzing phytoplankton cell counts and composition, water samples from each depth were fixed in Lugol’s iodine (1% w/v) and 3% formaldehyde and stored in dark until taken up for analyses. The number of phytoplankton present in all the grids was calculated. The total number of plankton present in 1 L of water sample was calculated using the formula.

\[ N = \frac{(n*v*1000)}{V} \]

Where, \( N \) = number of phytoplankton per Litre of water filtered
\( n \) = average number of phytoplankton in 1ml of plankton sample
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\[ v = \text{volume of plankton concentrate in 1ml} \]

\[ V = \text{total volume of water filtered in Litre} \]
Figure 2.3 (continued………….)

Asterionella sp.  
Coscinodiscus sp.  
Gyrosigma sp.

Melosira sp.  
Nitzschia sp.  
Navicula sp.

Peridinium sp.  
Closterium sp.  
Desmidium sp.

Spiragrya sp.  
Scenedesmus sp.  
Pediastrum sp.

Anabaena sp.  
Nostoc colony  
Oscillatoria sp.  
Phormidium sp.

Figure 2.3 (continued………….)
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Phytoplankton pigments

Determination of quantitative variation of phytoplankton pigments was done by spectrophotometric analysis (APHA, 1995). The chlorophyll a, chlorophyll b and pheophytin content of the extracts were determined by measuring optical density (APHA, 1995) using spectrophotometer. For the estimation of chlorophyll, the water sample was filtered through GF/C glass fibre filter paper, using a filtration apparatus fitted with vacuum pump. A thin bed of magnesium carbonate (MgCO₃) was applied to the filter paper. The filter paper containing the pigments were transferred to a clean beaker and added 5ml of 90% acetone, and the beaker was wrapped with aluminium foil, kept overnight at 4°C in a refrigerator. The contents were macerated and made up the extract solution to 10ml. The absorbance at wave lengths of 750, 665, 645, 630 and 450 nm of the resulting acetone. Pheopigments were measured by adding 2 drops of 0.5N HCl to the same sample and measurement of absorbance were performed at wavelengths 750nm and 665 nm. For the estimation of carotenoids, the above procedure was followed and the absorbance of the pigment extract was measured at wave lengths 510 and 480nm.
Estimation of primary productivity

Light and dark bottle method (APHA, 1995) was used for the estimation of primary productivity. The “Winkler” method of determining dissolved oxygen is normally used in the ‘light and dark bottle’ technique for studying production rates.

2.2.8 Secondary Production

Zooplankton

The samples were taken from subsurface water using standard net having mouth area 0.07m², 70 cm long. Maximum samples were collected at the cod end collecting tube. The sample which remained on the wall of the net was washed with water and transferred into the preservation bottles. The mesh size of 0.2mm of monofilament nylon is usually used for collecting zooplankton for taxonomic and productivity studies. In addition to the mesh size, the type, length and mouth area of the net, towing speed, time of collection and type of haul will determine the quality and quantity of zooplankton collected. The zooplankton collections can be made by horizontal, oblique and vertical hauls. In the horizontal sampling the net is towed at a slow speed usually for 10 minutes. The towing speed of the net should be such that the maximum amount of water enters through the mouth of the net for better filtration and gear used can withstand the strain. After fixation, the zooplankton are transferred and stored in airtight containers with sufficient quantity of preservative. While transferring, due care should be taken so that no part of the zooplankton sample is lost. The counting should be done under the microscope using Sedge wick rafter cell and when the specimen of a particular group is seen, a tally mark is made on the sheet. Zooplankton sampling and analysis were done following the
standard method (UNESCO, 1968; IOBC, 1969). Samples were preserved in 5% formalin (Goswami, 2004). Analysis of the zooplankton samples was done by group wise sorting. At first sub sampling was done after centrifuging the settled samples and the numerical abundance of each group in the entire 15 stations was computed and recorded.

2.2.9 Tertiary production

Fish Landing Centres

Data of fish landings of economically important food fishes adjacent to the sampling locations sited below were assessed seasonally from the following landing centres by stock assessment made through vendors.

1. Ernakulam market
2. Champakkara, Aroor
3. Chempu
4. Vaikom
5. Vypin
6. Varappuzha Jetty
7. Perumbalam

Gut content Analysis

The study of the feeding habits of fish and other animals based upon analysis of stomach content has become a standard practice (Hyslop, 1980). The specimens for gut content analysis were obtained from the respective sites during the sampling period. The fishes were brought to the laboratory, the specimens were dissected and their stomachs were dissected and their stomachs removed and preserved in specimen bottles containing 4% formalin. In the laboratory, total weights of the stomachs and stomach
contents were determined. Some of the stomachs were used to study food composition and selection. The stomachs were dissected and the content emptied into a petridish and they were washed well in 10 ml of water. The larger invertebrates such as chironomids and oligochaets, larvae were isolated counted and weighed. The rest of the material which constituted of detritus and plankton was thoroughly mixed with 10 ml of water in a beaker. A subsample was pipette out in a Sedgewick rafter cell which carries a volume of 1ml. The food items were then enumerated under a compound microscope. The analysis was made in triplicate. The occurrence of each food item was scored and then converted to a percentage by multiplying the ratio of the number of times an item occurred to the total number of guts analyzed by a hundred. The percentage abundance of each food item was also computed by multiplying the ratio of the number of a particular item in the stomach to the total number of items in the stomach by a hundred. The plankton samples taken from different locations where fish were sampled were identified and enumerated using a compound microscope. The organisms found in the food were identified to generic level and in few cases to species level wherever possible. The percentage occurrence and abundance of each item in the samples were analyzed in a similar way.

**Proximate composition of the selected fishes**

All biochemical analyses were carried out on fish samples previously oven dried at 60°C until constant weight and finely powdered with a pestle (Pulverisette2, FRITSCH).

**Total Proteins**

Proteins in water were hydrolysed with IN NaOH at 80°C for 30 minutes. Colour developed using Copper reagent and Folin-Ciocalteu
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reagent (Lowry et al, 1951). The absorbance was then measured at 750 nm using UV - Visible spectrophotometer.

**Total Carbohydrates**

Carbohydrates were estimated by the phenol-sulphuric acid method (Dubois et al., 1956). The samples were hydrolysed with IN conc. H$_2$SO$_4$ in 1: 1 ratio at 100°C for 1 hour and cooled at room temperature and filtered. To 1 ml of the aliquots, add 1 ml of 5% phenol and concentrated H$_2$SO$_4$, cooled the test tube and measured absorbance at 490 nm using UV Visible spectrophotometer (Genesys 10 UV).

**Total Lipids**

Lipids were extracted from the sediments and particulate matter according to the method of Bligh and Dyer, 1959. To 10 ml sample, 10 ml chloroform-methanol (2: 1 v/v) mixture and 20 ml of aqueous NaCl were added in separating flask and after thorough shaking. The preparation was allowed to stand for 30 minutes from the clean biphasic layer formed, the lower phase was removed and the same quantity of chloroform was added to make up the volume. This extract was dried in a vacuum desiccator, over silica gel and added 0.5 ml concentrated H$_2$S0$_4$, boiled maintaining in a water bath at 60°C. After cooling to room temperature, 5ml vanillin reagent was added and allowed to stand for 30 minutes. The absorbance of pink color developed was measured at 520 nm using spectrophotometer.

The fishes collected were virtually of the same size and sex as variability in size stands to affect the proximate composition and the mineral elements concentration. All the samples were collected fresh and refrigerated below 40°C prior use.
Moisture content

Estimation of moisture content was carried out by drying the pre-weighed wet samples at 60 °C until a constant weight was obtained. The difference in weight was calculated and expressed as percentage moisture content of the sample. Percentage was calculated by the following formula.

\[
\text{Moisture \%} = \frac{\text{Weight of tissue} - \text{Dry weight of tissue}}{\text{Wet weight of tissue}} \times 100
\]

The dried samples were finely powdered using mortar and pestle and stored in desiccators for further analysis.

Ash%

\[
\text{Ash\%} = \frac{\text{Weight of dry samples}}{\text{Original weight of the sample taken}} \times 100
\]

2.2.10 Statistical Analysis

Seasonal and spatial variations in water and sediment quality parameters were examined by Two way ANOVA without replication (Microsoft Excel, 2007). Pearson correlations analysis was carried out to find out the interrelations between different parameters (SPSS 13). PRIMER was used for statistical interpretation of data. In representative stations Multi dimensional Scaling (MDS) analysis was carried out to see species clustering / specificity using Plymouth routine in Marine Environmental Research (PRIMER- Clarke and Glorey, 2001). The main goal of the MDS is to detect meaningful dimensions underlying that allow the researcher to explain the observed similarities or dissimilarities between the factors, both biological and physico chemical. MDS helps to arrange plankton species abundance at different stations in a space so as to reproduce similarities. As a result it could explain the distances in terms of dissimilarity between species occurring in different stations. Technically it uses a function minimization algorithm that evaluates different
configurations with the goal of maximizing the goodness of fit or minimizing the lack of fit. Diversity is the precise expression of how individuals of a particular community are distributed in subsets of groups. To analyze changes in plankton community due to environmental influence relating seasons following diversity indices were also used.

**Species Richness (Margalef, 1968)**

Species richness is the number of different species in a particular area is calculated by

\[ D = (S-1) \log_e N \]

where S is the number of species and N is the total number of individuals of all the species in the sample.

**Species evenness (Heips, 1974)**

Species evenness is the relative abundance with which each species are represented in an area calculated by

\[ E = e \left( H(S) - \frac{1}{S-1} \right) \]

where \( H(S) \) is the species diversity in bits of information per individual and S is the number of species.

**Species diversity (Shannon and Wiener, 1963)**

The advantage of this index is that it takes into account the number of species and the evenness of the species. The index is increased either by having more unique species or by having greater species evenness.

\[ H(S) = -\sum [P_i \log_2 P_i] \]

where \( P_i = n_i / n \) (proportion of the sample belonging to the \( i^{th} \) species.)
References


