MATERIALS AND METHODS
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3.1 General Topography and Climate of Mysore district:-

Karnataka state with an area of about 21,600sq.kms extends between 74º 00' to 78 º 20' east longitudes and 11º45' to 18º 00' north latitude. Mysore district is situated in southern part of the Deccan peninsula and it forms the southern district of Karnataka state of India (Fig 3.1 and 3.2). The total area of Mysore district is 11,954 sq.km being sixth in rank among the district in the state in its size. Irrigation by canals is a characteristic feature of the district as the average rainfall is comparatively low. The climate of the district is moderate throughout the year.

Mysore district lies between 11° 39' and 12° 50' north latitude and 75° 45' and 77° 45'-east longitude. The soil of this region includes red fine loam to fine clay red to laterite type. The climate of the district is moderate throughout the year. It may be divided into four seasons. The pre monsoon from March to May followed by southwestern monsoon lashing up to the end of September. October and November may be termed as the post monsoon or retreating monsoon season. The period from December to February may be termed as dry season. The district receives major portion of its rainfall from the southwest monsoon. The normal annual rainfall is spread over a period of seven calendar months from the later half of April to October. The average annual rainfall in the district is 742.9 mm (Jan – Dec 2004) and most of the rainfall in the district is confined to the period from April to November. October is the wettest month. There is a meteorological observatory in the Mysore district. The period from March to May is on the continues rise in temperature. April is usually the hottest month with the mean daily maximum temperature 33.8º C and minimum at 21º C. Mysore is known as one of the garden cities of India and is also known thought the world for pomp and
gaiety of its traditional Dasara festival and fragrance of Jasmine. Cauvery river flows through the district. With important tributaries like Lakshmanthirtha, Kabini, Suvarnavathi, Shimsha, Hemavathi and Lokapavani on north. On south flows through the district and form draining and irrigating systems.

The district is bounded on the North by Hassan, Mandya district on the South. On the West by Kodagu district besides Wynad of Kerela state. On the East by Chamarajanagar. It can be classified as partly Maidan and partly semimalnad. Its physiography varies with an undulating land surface. The soil is fertile and well watered by perennial rivers, streams whole waters dammed by anicuts enrich their banks by means of canals. Here and there granite rocks arises from the plain which is otherwise un intermittent and wooded.

The name originated from the past as Mahisharashtra, Dipavamsha. Earliest records available is from 862 AD on Copper plate is Maysooru; later as Maisunad or Maisurnad is mentioned in the inscriptions of 11\textsuperscript{th} and 12\textsuperscript{th} centuries. As the literature flourished it is also spelt as Mahishurapura. The Mahishurapur or the buffalo headed monster that lived in this area and came to be killed by Chamundi. It has been ruled by Ganga’s, Pallava’s, Chola’s, Hoysalas, Wodeyar’s etc., Mysore district was divided into seven taluks for the purpose of administration namely H.D Kote, Hunsur, K.R.Nagar, Mysore, Nanjangud, Periyapatana and T. Narasipura. It is having 1203 villages and townships. The population according 2001 census is 262,249,11. Mysore is having good foresting composition since it lies in the southwestern part of Deccan plateau and stretches from the foothills of Niligiris in the south. The ecological factors are very diverse and hence different types of plant communities exist. Natural parks like Bandipur, Nagarhole ranges were established in 1974. The other wild life sanctuaries
like Biligirangana temple, Cauvery are rich is floristic composition. It is also rich in fauna. It has good large grazing animals, carnivores, birds, reptiles, mammals other groups of animals flourishing through out the district.

The importance is given to irrigation especially tank irrigation. The district is having 1221 tanks with canal irrigation systems, tanks, streams, springs, pools puddles and other water bodies where in algae can flourish very well. Keeping all these in mind, the sampling techniques and sampling stations have been selected. The district is having a combination of villages and town ships due to enrichment of towns. Due to industrialization and town ship programme, many numbers of villages and water bodies have been converted into residential areas. The green, watery areas have been converted into concrete tangles and tar roads. There is a lot of change in the microclimate occurring in this area.

The district is a tableland suited in the angle where eastern and western ghat ranges coverage into a group of hills called the Niligiri hills, which are lofty mountain ranges covered with vast forests. The general elevation of the district is more than 800 meters above MSL. Bettadapura hills in Periyapatna and Chamundi hills in Mysore. The western taluks receive more rainfall as compared to the portions of the district. The temperature ranges from 33°C to 16.8°C. The period from March to May is in continuous raise in temperature. April is the hottest month with temperature rising even upto 33.8°C; the day’s temperature drops during monsoon. December is the coldest month with mean daily maximum of 27.10 cg and minimum at 16.8 cg. The relative humidity is generally high during monsoon generally 88% and above during winter, relative humidity reduces to 30% during January and April (Fig 3.3).
Fig 3.1 - Physical map of India
Fig 3.2 - Physical Map of Karnataka
3.2 General Topography of the Study Site:-

(a) Shetty Lake

It is located at 12° 14’ 14” North longitude and 76° 39’ 37” East latitude at an attitude of 701.6 meters HMSL. It is situated 4 kms away from Mysore city, in the Mysore – Ooty road (Fig 3.3). It exhibits an arch shape. The water in it is used for agriculture and fishing with an independent catchment area of 1.79 sq.km; spread over an area of 4.8 hectares with a live capacity of 9 Mcft. The maximum depth of the lake when full is seven meters and has become heavily polluted due to continuous discharge of concentrated sewage, agricultural runoff as well as contaminated water from the neighboring Mandakalli lake which is also heavily polluted. It is a perennial lake. Floating vegetations like Eichornia, Lemma and Pistia are abundant. Aquatic vegetations like Typha, Potomogeton and Trapa are common along the edges. Luxuriant growth of all algal blooms throughout the year is well marked. The lake once harbored migratory birds but due to disturbances, these occasionally visit the lake (Plate 3.1a and 3.1b).

(b) Hadhinaru Lake

This lake is also called as Doddalake. It is located at 12° 02’ 02” North longitudes 76° 41’ 38” East latitude at an altitude of 653.35 meters HMSL. It is situated 33 kms away from Mysore city Lake is located in Nangangud taluk, which is 11 kms away from Nangangud town (Fig 3.3). It has an independent catchment area of 8.57 sq.km; with water-spread area of 10.10 hectares having a live capacity of 54.43 Mcft. The maximum depth of the lake when full is five meters. The lake bund has a top width of 10, front slope 1.1, near slope 2.1, and free board 3 meters. It receives water mainly from the kapila river and waruna cannel and the water is almost clean and is mainly used for irrigation. The lake harbors migratory birds during the seasons (Plate 3.2a and 3.2b).
Fig 3.3: Location of both lakes - A. Shetty lake and B. Hadhinaru lake
Plate 3.1a - Northern view of Shetty Lake

Plate 3.1b - Southern view of Shetty Lake
Plate 3.2a - Northern view of Hadhinaru Lake

Plate 3.2b - Southern view of Hadhinaru Lake.
3.3 Instrumentations: -

- **Thermometer**:

  0.0 to 100 HERMES BRAND mercury Thermometer was used to record the temperature of water at sampling sites and expressed as °C.

- **pH Meter**:

  pH was measured by using a digital pH meter model pH/mr meter OPH 14 INSTRUMENT TECHNIQUES PVT.LTD.

  Calibration of electrode was done with two standard buffer solution of pH 4.0 and 7.0 at 25 °C.

- **Colorimeter**:

  ERMA INC photoelectric colorimeter model AE – 11m. (4 -5, 2 Chome KAJICHO, CHIYODAKU, TOKYO, Japan) with OD value’s between 0 & 2 having the wavelength ranging from 420 nm to 660 nm with 1cm cell of 10nm light path was used for absorption measurements.

- **Electronic Digital Balance**:

  An electric digital balance ARCOSET MODEL 95 (THE BOMBARY, BURMBA TRADING CORP LTD,) with a sensitivity of 0.1mg was used for preparing standard solutions in the analytical works.
• **Camera Lucida Drawing Microscope:**

A combined Letiz Wet. Zar, Germany and Weswox optic and ERMA OPTICAL WORKS LTD. JAPAN was used for algae drawings.

• **BOD Incubator:**

INSREF, CAT NO. IRI, 27°C, 300 ml BOD bottles are placed in BOD incubator at 20°C for 5 days.

• **Secchi disk**

A Secchi disk is commonly used to measure the depth to which can be seen easily through the water, also called its transparency. Secchi disk is a circular disc of metal of 20 cm in diameter, painted alternately black and white in radial fashion. It has got a weight at the lower face so as to avoid a drift during lowering in water. A string is attached to it for lowering, which is marked in centimeters.

• **Miscellaneous Equipments:**

Pre-calibrated accurate pipette, burettes, measuring cylinder, beakers and conical flask. Test tubes were used during the analysis.

3.4 **Reagents and Chemicals:**

Best quality available, commercial samples of analytical grade reagents and chemicals were used and various concentration were prepared as per the Standard Methods, for the Examination of Water and Waste Water (APHA, AWWA, WEF 19Th edition 1995) and those described by Trivedy and Goel (1984); Chemical and Biological
Method for Water Pollution Studies. Also certain methods prescribed in Indian Standard Method (ISI, 1982) were also employed.

3.5 Physico-chemical Analysis:

- **Temperature:**
  
  Mercury thermometer (APHA 1995, 2-59 pp)

  Atmospheric and water temperatures were recorded at sampling stations and water temperature measured at a depth of 12 cm. with the help of Mercury thermometer $^\circ C$. (Is 3025)

- **Hydrogen Ion Concentration (pH):**


  pH was determined with the help of Universal pH paper at the sampling sites and later confirmed in the laboratory by using digital pH meter equipped with a calomel reference electrode and a glass electrode.

- **Dissolved Oxygen (DO):**

  *Winkler’s Iodometric method* (APHA-1995, 4-98 pp)

  Samples were fixed in a 300 ml BOD bottles. DO was used to oxidize manganese ions to manganese ions. With the help of an alkali (KOH). Then a strong acid ($H_2SO_4$) was added to reduce magnetic ions with the help of iodide ions and in the process iodine equivalent to the original concentration of DO. (in the sample) was liberated. This iodine was estimated by titration against standard Sodium thio sulphate using starch as an indicator.
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- **Biological Oxygen Demand (BOD):**

*Standard methods* 5 day BOD (APHA-1995, 4-98 pp)

The samples were incubated for 5 days at 20 °C with the necessary preconditions (dilution, addition of nutrients and neutralization). The sample was filled into two BOD bottles. One bottle was kept in BOD incubated for 5 days at 20°C. The DO content in another bottle was determined immediately in the laboratory with modified Winkler’s method and taken as initial DO. After 5 days of incubation the D.O. in the bottle was determined and it as D-5. BOD was calculated using the formula.

\[
\text{BOD, mg/l} = (\text{DO-D}_5) \times \text{Dilution factor}
\]

DO=initial D.O. in the sample

\(D_5 = \text{D.O. after 5 day's of incubation.}\)

- **Total Alkalinity:**

*Standard methods* (APHA, 1995, 2-25 pp)

To estimate total alkalinity titrimetric method was employed. 100 ml of sample was titrated against 0.1N Hydrochloric acid until the color disappears at the end point using phenolphthalein indicator at end point. This is phenolphthalein alkalinity; 2 or 3 drops of methyl orange were added to the same sample and the titration continued until the yellow color changes to pink at the end point, this is total alkalinity (This includes Carbonates and Bicarbonates).
• **Total Hardness:**

*Standard methods* (APHA-1995, 2 - 36 pp)

50 ml sample was titrated against EDTA (0.01N) using Erichrome black-T as an indicator to a blue end point and expressed as mg/L.

• **Calcium (Ca):**

*EDTA Titrimetric method* (Standard methods APHA-1995, 3-50 pp)

Calcium hardness was estimated by using EDTA with sodium hydroxide, with muroxide as an indicator until the pink color changes to purple and the values expressed as mg/L.

• **Magnesium (Mg):**

*Calculation method* (Standard methods APHA-1995, 3 - 75 pp)

Magnesium was calculated from Magnesium hardness, which was obtained by deducting the value of calcium hardness from total hardness, and expressed as mg/L.

• **Chloride (Cl):**

*Argentometric titration method* (Standard methods APHA-1995, 4 - 49 pp)

50 ml of the sample was titrated against 0.01N silver nitrate using potassium chromate as an indicator to a brick red end point. Silver chloride is precipitated quantitatively before red silver chromate is formed and the value expressed as mg/L.
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• **Carbonates (CO$_3^{2-}$):**

  *Standard methods (APHA - 1995)*

  Carbonate was estimated by titrating the sample with a strong acid like HCl or H$_2$SO$_4$, 500 ml of the sample was titrated against N/20, H$_2$SO$_4$ using phenolphthalein as an indicator to a colorless end point and expressed as mg/L.

• **Bicarbonate (HCO$_3^{2-}$):**

  *Standard methods (APHA - 1995)*

  50 ml of sample was titrated against 0.1 N Hydrochloric acid by using methyl orange as an indicator which turns pink at the end point and expressed as mg/L.

• **Phosphates (PO$_4^{3-}$):**

  *Standard methods (APHA-1995, 4 - 112 pp)*

  It can be determined as inorganic orthophosphate. Phosphates react with ammonium molybdate to form molybdophosphonic acid, which is reduced to a blue color on addition of stannous chloride. This blue color can be measured at 690 nm using calorimeter to calculate the concentration of phosphates.

• **Sulphate (SO$_4^{2-}$):**

  *Gravimetric Method with Drying Residue (APHA-1995, 4 - 177 pp)*

  Sulphate is precipitated as BaSO$_4$ in HCl medium by addition of BaSO$_2$ Solution. The reaction is carried out near the boiling temperature. The precipitation is filtered, washed to remove the chlorides, dried or ignited and weighed as BaSO$_4$. 

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• **Nitrate (N0<sub>3</sub>\(^-\))**: 

*Bruicine- sulphate method* (Standard methods APHA-1995, 4 - 159 pp)

Nitrites react with bruicine sulphate and sulphanilic acid to form a nitrodervative, which in alkaline media develops yellow colour due to rearrangement of its structure. 10 ml of H<sub>2</sub>SO<sub>4</sub> is added until it turns brown colour. It is boiled in a bath and cooled until it develops a yellow colour. The colour developed was read in a colorimeter using at 410 nm. Potassium nitrate was used as the standard. The results were expressed in mg/L.

• **Nitrite (N0<sub>2</sub>\(^-\))**:  

*Colorimetric Method with Drying of Residue* (APHA-1995, 4 – 177 pp)

Nitrite forms a diazonium salt with sulphanilic acid medium (2.0 – 2.5 pH), which combines with a – naphthlyamine hydrochloride to form a pinkish dye, which can be determined Colorimetrically.

• **Total phosphorous**:  

*Digestion method* (APHA-1995, 4 - 151 pp)

All forms of phosphorous, whether dissolved or particulate are converted to inorganic forms (Phosphate) after digestion or oxidation of the sample. H<sub>2</sub>SO<sub>4</sub>, K<sub>2</sub>SO<sub>4</sub>, H<sub>2</sub>SO<sub>4</sub>, HNO<sub>3</sub> technique is employed for digestion of sample and the phosphate thus released can be determined colorimetrically. The concentration of phosphorous is obtained from the standard curve and multiplied it by dilution factor.
• **Secchi Transparency:**

*(Trivedy and Goel, 1984, 42 pp)*

\[
\text{Secchi disc light penetration} = A + \frac{B}{2}
\]

Where A = depth at which disc disappears  
B= depth at which secchi disc reappears

• **Chlorophyll a:**

*Acetone method* *(Trivedy and Goel, 1984, 126 - 127 pp)*

Chlorophyll a determined using the acetone extraction method and measured using spectrophotometer. Absorbance is made at 630nm.

• **Total Dissolved Solids (TDS):**

*Evaporation methods* *(Standard method APHA-1995, 2 - 58 pp).*

100 ml-filtered samples were evaporated on a water bath in pre-weighed porcelain dish; and final weights of the evaporation dish were recorded using the analytical balance and results expressed as mg/L.

3.6 Estimation of Phytoplankton’s:

Water samples were collected from the lakes for plankton analysis in black colored plastic carboys of one liter. Filamentous algae and other floating debris were avoided. For each sample collected, 25 ml of 4% formaldehyde was added *(Welch, 1948)* with few drops of Lugol’s iodine. Sedimentation was done in glass columns. The sediment was finally reduced to 20 ml and was preserved in a glass vial. From each vial one drop was mounted on a slide and a cover slip was carefully put over it. Five high
power fields (15X x 45X), one in each corner of the coverslip and are at the center were made and the algal populations were estimated. These observations were at random and were repeated four times for each sample. This procedure was repeated for each sample and the number of each organism was extra plotted to extract number of organism/L (Rao, 1995). Identification of plankton to species level were made by camera lucida drawings/photographs using monographs and research articles and expressed as number of organisms/L.

3.7 Algal Enumeration:

The samples were collected from both lakes for 12 months; Sedimented and collected in 30 ml vials. The samples have been collected in 30 ml vials. To these samples few drops of Lugol’s iodine and few drops of 4% formaldehyde was added and labeled. The preserved samples were observed under microscope. Camera lucid drawings with measurements were written. Phytoplankton has been identified up to the species level by using monographs and research articles. Photographs of some important taxa were also made and presented in the thesis.

The algae from two lakes have been identified and listed. A key with taxonomic description up to species level has been prepared. The physico-chemical, biological data of two represented lakes were used for ecological considerations of the waters and the trophic status of waters has also been discussed using statistical analysis. Few of indexes like Nygaard’s index, Shannon and weaver diversity index, Dominance, frequency distribution of species have been discussed.
Phytoplankton count was done by Lackey’s Drop Method (1938) as mentioned in APHA (1995) and modified by Saxena (1987).

Formula used for the calculation of phytoplankton as units/l is

\[
\text{Phytoplankton Unit/L} = \frac{n \times v \times 1000}{V}
\]

\(N=\) No. Of phytoplankton counted in 0.1 ml.concentrate.

\(C=\) total volume of concentrate in ml.

\(V=\) total volume of water filtered through net

3.8 Statistical analysis:-

The data obtained from physico-chemical analysis and biological analysis was subjected to Pearson’s correlation matrix. The Dendrogram and Pearson’s correlation matrix are obtained by Statistical Package for Social Science (SPSS - Software package.10.05 version). Principle Component Anaysis (PCA) was done. This data provides a brief but a precise explanation of the interrelationship of the various physico–chemical and biological parameters analysis. Data can be analyzed by multivariate analysis, PCA scatter biplot are obtained from PASTA software package.