Chapter III

MATERIALS AND METHODS

Study area:

The present study was carried out in the North Guwahati area of Kamrup district in Assam. The North Guwahati area is the northern most part of the city Guwahati. The region is situated at the northern bank of the river Brahmaputra in central Assam, India. The entire area of the region is about 52 square kilometers and at a height of 168 feet above from the sea level. The coordinate of the region is located at 26°10' - 26°20' N and 91°40' - 91°50' E. The region has a sub-tropical climatic condition. It experiences warm, humid summers and mild winters. The area has an annual average humidity of 81.01 percent. The annual average temperature recorded in the region is 28.67°C. The area receives an annual average rainfall of 159.7 cm. The area is unevenly distributed with water bodies throughout. The region is connected with the Brahmaputra River by drainage like 'Nowakhalijaan', 'Ghorajaan', 'Siligurijaan' etc. The region is gradually developing its appearance into an urban scenario and would develop into an industrial city within a decade. The place holds many fish ponds either formed naturally or artificially where algal ecology is needed to be explored.

Study site:

In the present endeavor, four water bodies, i.e. fish ponds were selected from North Guwahati area, Kamrup, Assam. We also designated the study sites as S1, S2, S3 and S4. The study sites are described below in **Table 1**:

Sites	Latitude	Longitude
S1	26º15'41" N	91º41'52" E
S2	26º15'23" N	91º41'35" E
S3	26º15'34" N	91º41'36" E
S4	26º15'29" N	91º41'41" E

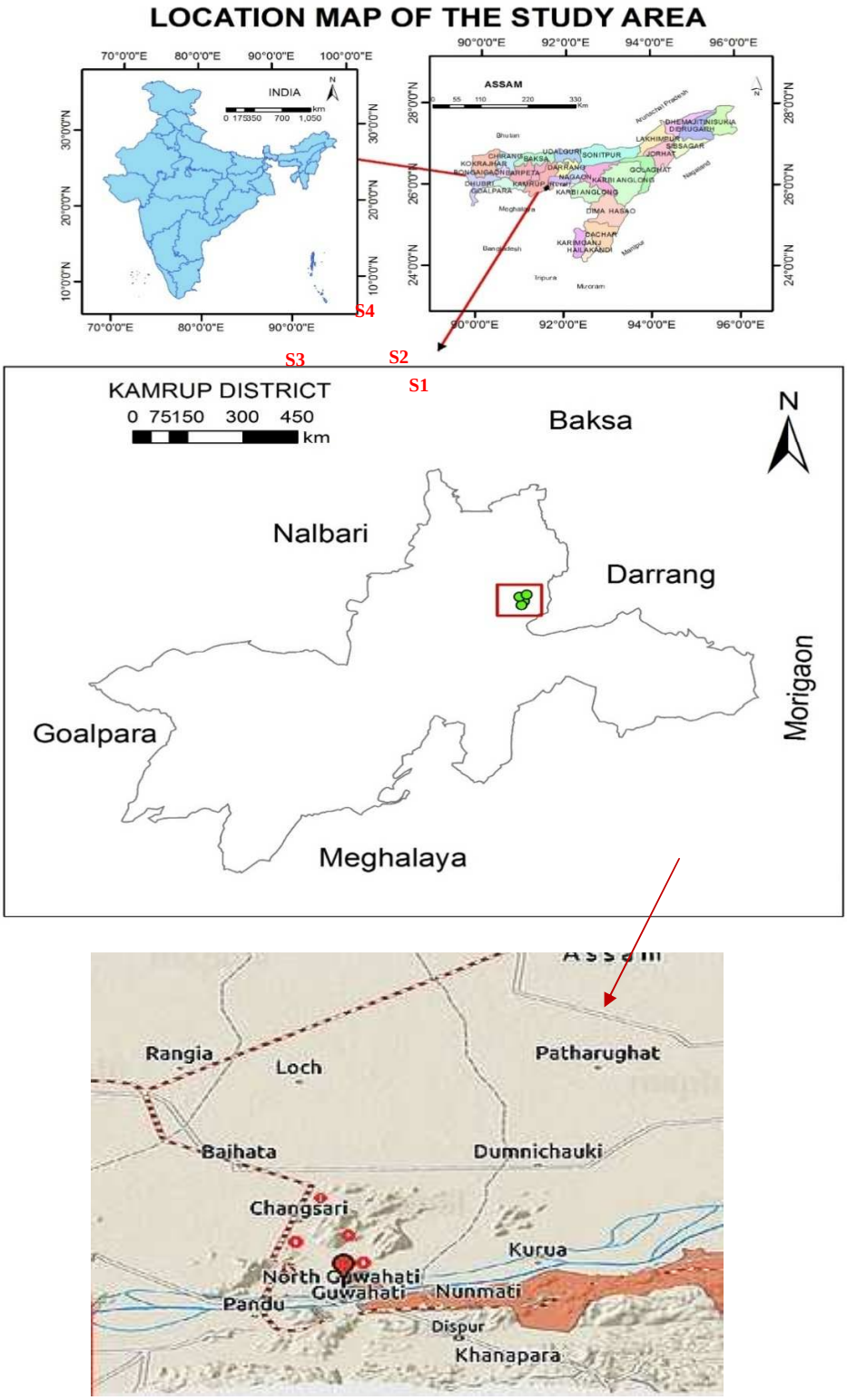

Fig. 1: Map of the Study area

The fish ponds selected as study sites for algal exploration are all located within human habitats and experiences maximum human intervention and disturbances. Local villagers use those ponds to culture fish and clean the ponds occasionally. The water bodies also experience pollution from the nearby human habitation.

Study site S1 is a public fish pond near a road side Namghar (Place of worship). Study site S2 is a pond near a Water Treatment plant. This pond rarely witnesses pollution being located at a distance from the town area and remains mostly clean. The study site S3 is another fish pond owned by some local villagers. And, the study site S4 is located quite near to the railway track in a recently developed township.

METHODS OF SAMPLING:

The specimens were collected at a monthly interval from April 2018 to March 2019 for a period of one year. The sample collection was done in the morning hours between 6 am to 10 am. Water samples were collected at random stations from the surface level of each of the ponds. The samples were collected using Nansen sampler.

COLLECTION OF ALGAL SAMPLES:

The water samples for micro algal analysis were collected by filtering techniques with the help of plankton net of bolting silk whose mesh size was 45µm. Almost 10 liters of water was allowed to pass through the mesh and the remaining cellular materials were then transferred to clean plastic bottles. For cases, when planktons having sizes less than 45µm, water samples were directly collected in separate water bottles. The visible algal samples were also collected with forceps from side of the ponds by hand picking method. After collection, the bottles containing

algal samples were properly labeled and brought to the laboratory of Department of Botany, Gauhati University for further analysis to be carried out.

Ecological notes of the habitat, pH, temperature and conductivity of the water sample were recorded in a field note book in details. From the collection spots, digital photographs were also taken, which shows algal representatives in their natural condition and habitat.

The collected samples were preferred to be observed in fresh condition. Sometimes the collected algal samples were left overnight to get concentrated by gravimetric method. For preservation purpose, to prevent it from decay and degradation, the remaining samples were fixed in Lugol's Iodine solution following Das and Keshri (2013).

MICROSCOPIC OBSERVATION:

For observation of the fresh algal samples, a drop of algal sample on a microscopic slide was mounted with a cover slip upon it carefully. The slide was then observed using Euromex Delphi-X Observer series microscope with Image View software under proper and different magnifications and digital photographs were taken. Measurements were taken using micrometer scale.

IDENTIFICATION OF SAMPLES:

Identification of the algal samples were done by consulting standard literature and monographs of Desikachary (1959), Prasad and Srivastava (1992), Prasad and Mishra (1992), Gandhi (1999), Whitton and Brook (2005), Perumal and Anand (2009) and Yamagishi (2010), Das and Adhikary (2014), Das and Keshri (2016).

WATER QUALITY ANALYSIS:

- **Surface water temperature:**

  Soon after the collection of the water samples, the surface water temperature was measured on spot in a 500ml capacity beaker. Systronics digital water analyzer 371 was used for the measurement. To minimize the errors, the instrument was calibrated prior to use with a thermometer of known accuracy and is expressed in degree Celsius, designated as " ^{o}C ".

- **pH:**

  The pH of the water samples were also determined on spot. Systronics digital water analyzer 371 was used for the measurement. To calibrate the electrode of the instrument, Standard buffer solutions were used.

- **Conductivity:**

  The measurement was carried out using Systronics digital water analyzer 371. Conductivity was recorded as micro siemens per centimeter, represented as µS/cm.

- **Sorenson similarity coefficient:**

To measure the similarity between algal communities of two study sites, Sorenson similarity coefficient was used. This simple similarity measure deals only with presence-absence data of the algal species. Botanist Thorvald Sorensen (1948) developed it. Its value always ranges between 0 to 1. Sorenson similarity coefficient is calculated by using the given formula:

$$Ss = \frac{2C}{A + B}$$

Where, Ss= Sorenson similarity coefficient

A = Number of plant species in community A

B = Number of plant species in community B

C = Number of plant species common to both the species.

❖ **Palmer's pollution index:**

For detection and evaluation of water pollution, water pollution indices like species composition, diversity of species, their distribution pattern, the presence or absence of indicator species or their group etc. may be used as indicator to estimate pollution status. Palmer (1969) took the initiative in this regard, identified and arranged a list of 60 genera and 80 species of algae that are tolerant to organic pollution. This index table included 20 algal genera most tolerant to organic pollution and also their respective score (or number) depending on their relative tolerance. The numbers scored by each genus present in a sample are totaled to get the Palmer's pollution index of the water body.

Sl. No	Genera	Pollution Index	Sl. No	Genera	Pollution Index
1	*Anacystis(Microcystis)*	**1**	11	*Micractinium*	**1**
2	*Ankistrodesmus*	**2**	12	*Navicula*	**3**
3	*Chlamydomonas*	**4**	13	*Nitzschia*	**3**
4	*Chorella*	**3**	14	*Oscillatoria*	**4**
5	*Closterium*	**1**	15	*Phormidium*	**1**
6	*Cyclotella*	**1**	16	*Pandorina*	**1**
7	*Euglena*	**5**	17	*Phacus*	**2**
8	*Gomphonema*	**1**	18	*Scenedesmus*	**4**
9	*Lepocinclis*	**1**	19	*Stigeoclonium*	**2**
10	*Melosira*	**1**	20	*Synedra*	**2**

The Palmer pollution indexes of algal genera are:

Palmer (1969) formulated the following pollution index scale for assessment of organic pollution of water body on the basis of the above table:

Pollution index	Pollution Status
< 15	Very light organic pollution
15 – 20	Organic pollution
> 20	High organic pollution