CHAPTER I: INTRODUCTION

The diatoms are one of the prominent and ecologically most significant groups of organisms on the earth. They are unicellular, eukaryotic, photosynthesizing algae, having a siliceous skeleton and are found in all aquatic environments. The division Bacillariophyta is illustrious by the presence of an inorganic cell wall composed of silica (hydrated SiO_2). The wall or frustule consists of two parts called "valves". Diatoms have evolved to have detailed silica cell walls that reflect the types of habitat to which the particular species is adapted. Diatoms multiply rapidly, maintaining a dynamic population of varying size. They range in size between 20-200 microns in diameter or length in size, although sometimes they can be up to 2mm long. The cell is solitary or colonial. They are non-motile or capable of only limited movement along a substrate and play ecological roles in producing diatomaceous earth and toxic blooms in surface waters.

Among unicellular microalgae, diatoms probably represent one of the most diverse groups with the number of species estimated to be between 10000 and 100000 (Round et al 1990). Diatoms may occur in such large numbers and be well preserved enough to form sediments composed almost entirely of economic benefit being used in abrasives , filters , paints, toothpaste and many other applications . Fossil evidence suggests that diatoms originated during or before the early Jurassic period.

Rivers form a lifeline associated with people community. Rivers engage in a vital function throughout assimilating or perhaps carrying industrial and municipal water and runoff from agricultural fields. Now a days, rivers are generally amongst the most susceptible water bodies to pollution on account of unprecedented progress. Consequently, the water quality of these water resources is an issue tied with ongoing matter and it has contributed to a rising dependence on assessment of river water quality. The standard of water is actually followed by its physical, chemical and biological features.

Diatoms are popular tool for monitoring environmental conditions of past and present and are commonly used in studies of water quality. They are widely used as assessment tools in understanding and management of environments. Diatoms-based indices are increasingly becoming important tools for assessment of environmental conditions in aquatic systems. Several diatom indices are tested in this study for application in the Cauvery River.

In 21st century, discussions of diatoms are everywhere as the roles they play in global nutrient, oxygen and silica cycling, their utility in understanding the status and trends of aquatic ecosystem health, harmful algal blooms and also in their potential as a possible source of renewable fuels. Diatoms often have narrow windows of tolerance to pH and nutrients which have been widely studied and defined. Diatoms have proven to be extremely powerful indicators with which to explore and interpret many ecological and practical problems related to health sciences, forensic sciences, nanotechnology and global environmental changes (Smol and Stoermer, 2010).[15]

Diatoms have been used in forensic science in a variety of ways, the most frequent being the diagnosis of death by drowning. When a person drowns, water will enter the lungs and then enter the bloodstream through ruptures in the peripheral alveoli before being carried to the other organs such as liver, heart etc. Diatoms do not occur naturally in the body. If laboratory tests show diatoms in the body that are of the same species found in the water where the body was recovered, then it may be good evidence of drowning as the cause of death. Based on the study of drowning victims, where the diatoms are present in the medium, the penetration of diatoms into the alveolar system and blood stream has been caused by the breathing in of water by the drowning victims and then leads to the penetration of diatoms into other organs and parts of the body, such as bone marrow, the brain, kidneys, and lungs. Hard bones (sternum and femur) and soft tissues (lungs and liver etc) of drowned bodies are usually sent to the Forensic Science Laboratories for the detection of diatom. While solving drowning cases, a correlation between the diatoms extracted from these tissue samples and the samples obtained from putative drowning medium has to be established for the successful determination of drowning site. The occurrence of diatoms in the bone marrow is a proof that the individual was alive when entered the water. This means that the cause of death was due to the drowning. More suggestions were given by that diatom test would be of much importance in the diagnosis of drowning cases, origin of diatoms found in bone marrow is known i.e. matching of diatoms from both putative water medium and tissue of drowned body is must require for the success of this test. Some previous important reviews on diatoms and drowning have been published by Holden and Crosill, Ruston, Timperman, Peabody. Among the various works have made good efforts to make this study live and hopeful one.[11]

CHAPTER II: LITERATURE REVIEWS

A Kock et.al (2019) studied the diatom community structure and relationship with water quality in Lake Sibaya, KwaZulu-Natal, South Africa. The study aimed to determine the diatom community structure and it's response to changes in water quality. Water and diatom samples were collected from four sites in Lake Sibaya during August 2015, December 2015 and February 2016. All samples were collected and analysed according to standard methodologies. From the results it was clear that the Lake had a lower diatom diversity than that described in a previous study in 1966 with cocconeis plancentula, Gomphonema species, Epithemia adnata and Nitzschia species being the dominant taxa.[1]

Guangtao XU et.al (2011) studied the application for drowning identification by Planktonic diatom test on Rats. They established a model of drowning and by investigating diatoms in Lung, Liver, Kidney and long Bone marrow of rats at different time to discuss the cause of death. The organs of 35 rats were extracted 0.5 h, 1 h, 6 h, 12 h, 24 h and 48 h after drowning and the of sham-drowning group killed by mechanical asphyxia were extracted 1 h after body immersed in water. The organs were digested by acid and the diatoms were analysed by statistics. Result shown the detection rate was 100% 6 hours after drowning except the sham-drowning group, just only one case was positive in the lung. So it is concluded that the detection rate of diatoms could be considered as important evidence drowning determination.[2]

J V Patile et.al (2013) studied the seasonal variation of diatoms density and species richness of Lotus Lake, Toranmal(MS) India. This reveled that the density of diatoms was maximum in summer, while minimum species richness was recorded in winter. The diatoms structure depends on a variety of environmental factors that include biological parameters as well as various physic-chemical factors. Mean Standard Error of Mean (SEM) was calculated for each season and one-day ANOVA with no post test. The Pearson correlation was calculated by keeping diatoms as dependent variables and other abiotic factors as independent variables.[3]

Sarika Grover et.al (2017) recorded the Eco-Taxonomical studies on diatoms from the Chambal River (CENTRAL INDIA). A checklist of diatom taxa has been prepared for the Chambal River which includes 102 samples sites along the river. Most of the diatom taxa belong to classes Bacillariophyceae followed by Fragilariophyceae and Coscinodiscophyceae. Dominant diatom species belong to genera like Cyclostephanos, Cyclotella, Melosira etc.[4]

Ruth E Holland (1980) studied the seasonal fluctuations of major diatom species at five stations across Lake Michigan. From 27 May, 1970 to 4 October, 1972, they collected water samples from five stations on a transect across Lake Michigan from Milwaukee, Wisconsin to Ludington, Michigan. At all stations in the 3 years sampled, diatoms reached their highest numbers in late spring or summer. These periods of greatest abundance of total diatoms were the result of the rapid growth of one or two species. Stephanodiscus hantzschii was the dominant species or one of the predominant forms at the other 4 stations. Cyclotella stelligera was the dominant diatom when highest yearly numbers were reached at Stations 3 and 5 in August, 1972.[5]

P C VOS and MISDORP (1985) studied the sediment stabilization by Benthic diatoms in the Intertidal area of the Ooster-Schedule. Investigations in the laboratory (Holland ea, 1974) and in the field (Coles, 1979 and De Boer,1981) show that microphytobenthos can be an important factor in stabilizing intertidal sediment surfaces. Especially benthic diatoms improve the stability of the sediment by secreting mucilaginous acidicpolysaccharides o the sediment stability activity of benthic diatoms depends upon the density of benthic diatoms, and the mucilage secretion of the various species.[6]

Minchen et.al (2019) determine how the distribution of diatoms in surface sediments on the inner shelf of the East China Sea varied by season and because of a Typhoon, by examining samples collected in 3 surveys, one in December 2008 and 2 in August 2009, before and after a typhoon. They identified 64 species and varieties of diatoms from 27 genera in the samples. They identified 5 diatom assemblages that represented the different Oceanographic conditions in the study area, the spatial distributions of which were closely related to the environmental conditions. The distribution of diatoms in surface sediments in the study area were modified considerably by Typhoon Morakot, which suggests that the diatoms seasonal cycle is completely disrupted because of typhoons that mainly occur in August and September.[7]

Maria Helena Novais et.al (2019) studied the vertical distribution of benthic diatoms in a large reservoir during thermal stratification. During the Alqueva hydro-meteorological Experiment (ALEX) field campaign, diatom communities were studied in the margins and in 3 platforms (from the surface to the bottom of the reservoir) located in the limnetic zone of the Alqueva reservoir, one of the largest artificial lakes in western Europe. A detailed meteorological and physico-chemical characterization of the reservoir was carried out from June to September in summer 2014, when the reservoir was stratified, to relate these variables with diatom assemblages. Taxa richness Shannon diversity Index, Pielou's evenness and Specific Pollution Sensitivity Index (SPI) also differed with depth, with the lowest values of all indices detected at surface samples, increasing with depth, reaching the highest values at 20m for taxa richness, Shannon diversity and pielou's evenness indices.[8]

Bertrand Ludes et.al (1999) studied the diatom analysis in victim's tissue as an indicator of the site of drowning. The diagnosis of drowning is one of the most difficult in forensic pathology and previously we proposed criteria for a positive tissue analysis according to the qualitative and quantitative diatom investigations. In the positive cases, they studied the reliability of determining the site of drowning by comparing the diatom taxa found in the lung samples with those of the water samples or in the absence of these samples with the result of the water diatom monitoring program set up in their region. The result showed that a concordance of the abundance of the diatom taxa in tissues compared to the site of drowning and their distribution relative to one another was 65% in the group where the site of drowning was known and 35% in the other group. The concordance of the individual distribution in the lungs of water diatom taxa may be an interesting method to guide the investigations for determining the site of drowning. The two limiting factors are the concentration of diatoms in the lungs and the development of a river monitoring program in the district of the study.[9]

Ruth Patrick (1973) studied the use of Algae, Especially Diatoms in the assessment of water quality in biological methods. Two main systems of approach used to determine if algae can reliable indicate water quality are discussed in this paper. One approach is to observe and analyze natural communities. The effect of a pollutant can be estimated by shifts in species composition and structure of the community in this type of study. The second approach studies a single or a few species in cultures in the laboratory under known and carefully regulated conditions. These studies are valuable in determining the physiological and morphological changes in function rates and polymorphism due to concentration of a given chemical or physical factor.[10]

CHAPTER III: AIM AND OBJECTIVES

Aim

To determine the different species of diatoms in various water resources in the area of North Paravoor region Kerala.

Objectives

- To extract the diatoms from different water samples by using Acid Digestion method.

CHAPTER IV: MATERIALS AND METHODOLOGY

Materials:

Apparatus:-

1. Conical flask,
2. Samples
3. Centrifuge
4. Beakers
5. Micro pipette

Chemicals:-

1. Sulphuric acid
2. Nitric acid
3. Ethanol (for the preservation of diatoms)

Instruments:-

1. Magnus MLX Plus Compound Microscope
2. Smart R17 Plus Mhanil Centrifuge

Figure 1: Compound Microscope

Figure 2: Centrifuge

Methodology

100 ml of water samples were collected from three different water resources in North Paravoor region. 3 water samples from each three locations of the three different water resources were collected. The water resources are Chennamangalam River, Thattukadavu River and Chittattukara Stream. On 1st January 2020, started the method of Acid Digestion for the identification and extraction of Diatoms. 50 ml of the water sample is mixed with 25 ml of concentrated Nitric Acid. Then make it rest for 48 hours in a dark room, like that 9 water samples from the Chennamangalam River were prepared. Then on 3rd January 2020 took the water samples for microscopic examination. 1.5 ml of the water sample is took in a tube by using the micropipette, like that make other 8 water samples also and centrifuge it for 10 minutes. After 10 minutes remove half portion of 1.5 ml solution and add the same amount of distilled water to it. Then again centrifuge it for 10 minutes. After that, took the sample in a glass slides and make it air dry. Then observe the slides under the compound microscope and took the photographs. On that day only 9 water samples from the Thattukadavu River in morning and 9 water samples from the Chittattukara Stream in afternoon were prepared for the Acid Digestion method. And the same microscopic examination process were repeated on 5th January 2020.

Figure 3: Sample beaker

Figure 4: Samples in the centrifuge machine

CHAPTER V: OBSERVATIONS

Chennamangalam River

Location-1:

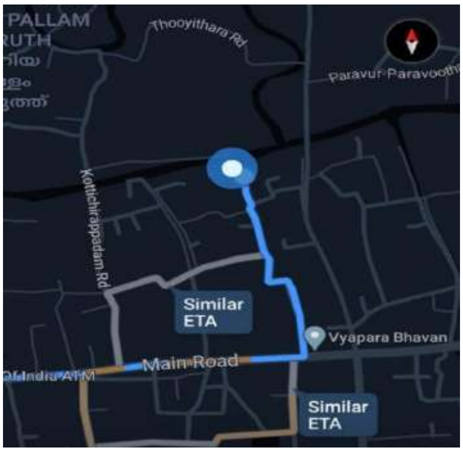

Figure 5: Location 1 Chennamangalam River

Figure 6:Sample Slide 1

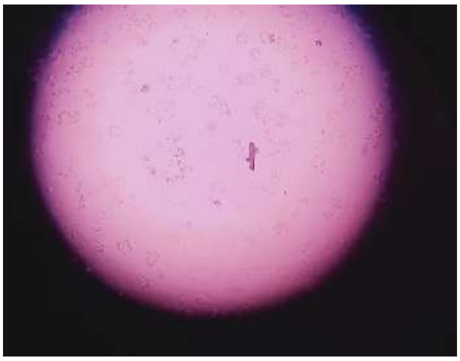

Figure 7:Sample 1 Pinnate Diatom

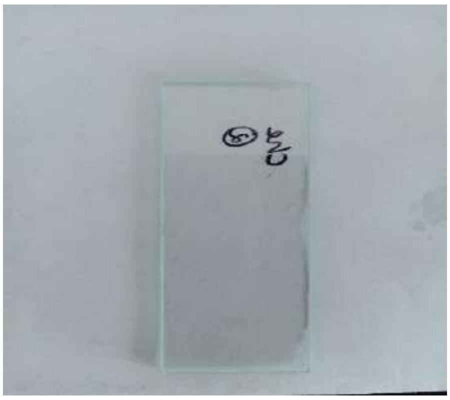

Figure 8: Sample Slide 2

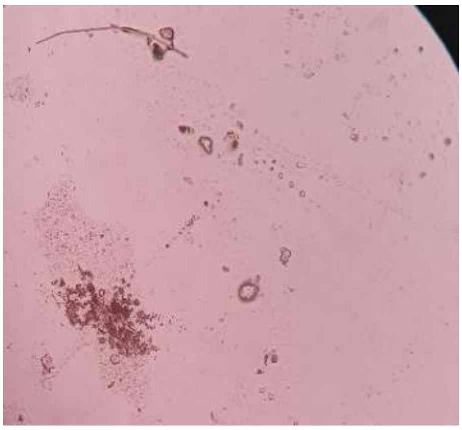

Figure 9: Sample 2 Centric Diatoms

Figure 10: Sample Slide 3

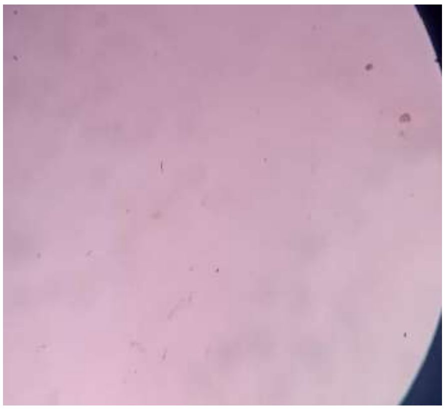

Figure 11: Sample 3 Centric and Pinnate Diatoms

Location 2:

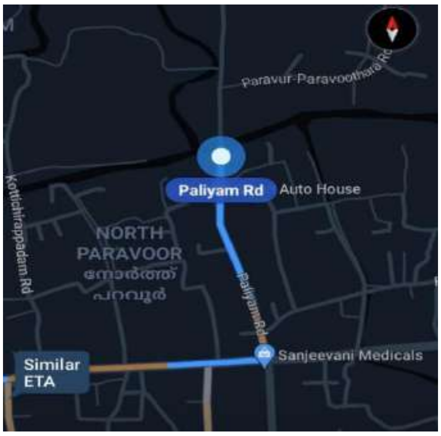

Figure 12: Location 2 Chennamangalam River

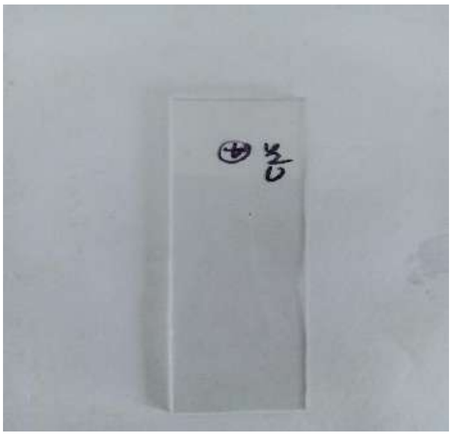

Figure 13: Sample Slide 4

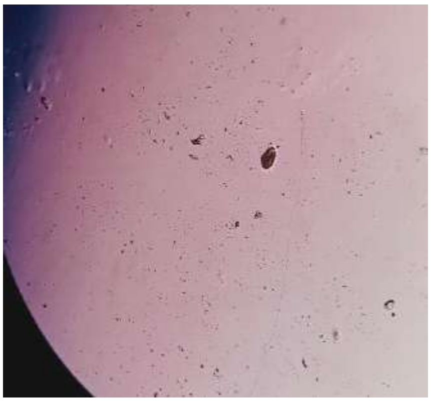

Figure 14: Sample 4 Centric Diatoms

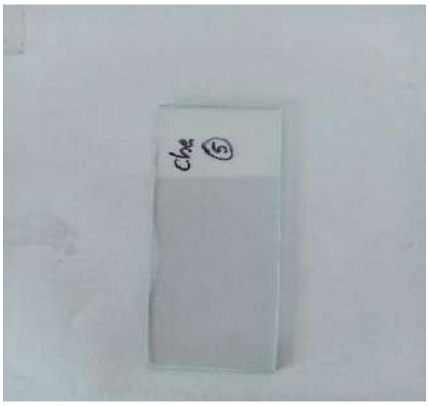

Figure 15: Sample Slide 5

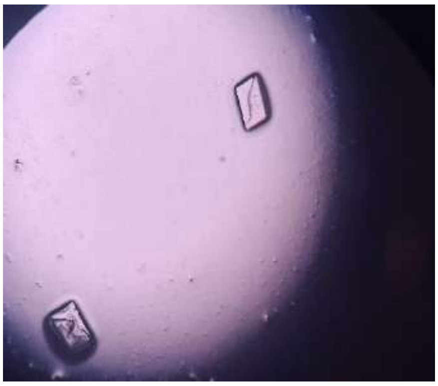

Figure 16: Sample 5 Centric Diatoms

Figure 17: Sample Slide 6

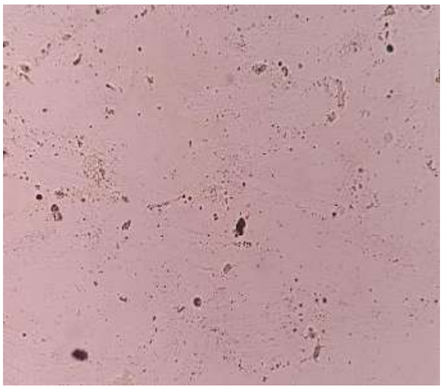

Figure 18: Sample 6 Centric and Pinnate Diatoms

Location 3

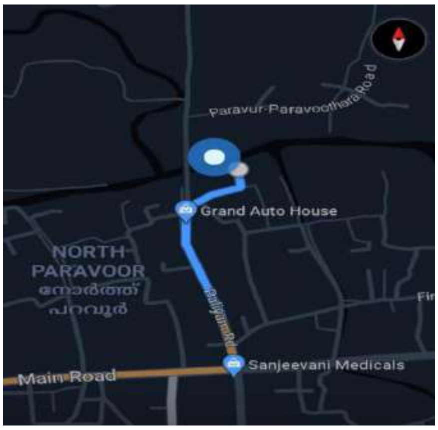

Figure 19: Location 3 Chennamangalam River

Figure 20: Sample Slide 7

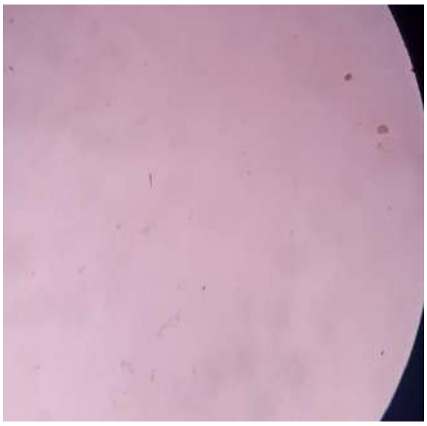

Figure 21:Sample 7 Centric Diatoms

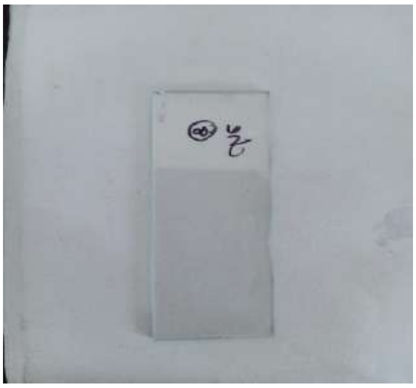

Figure 22: Sample Slide 8

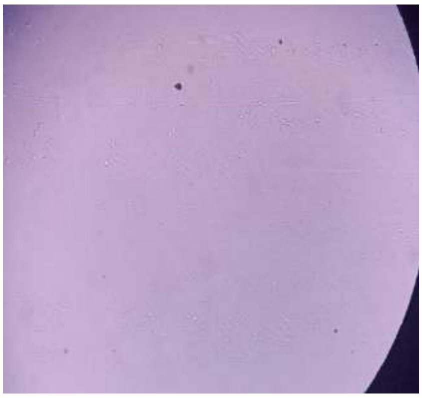

Figure 23:Sample 8 Centric Diatoms

Figure 24: Sample Slide 9

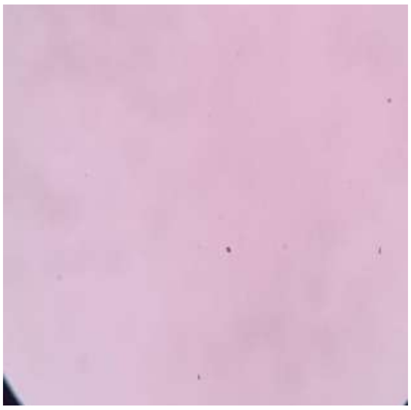

Figure 25: Sample 9 Centric Diatoms

Thattukadavu River

Location 1

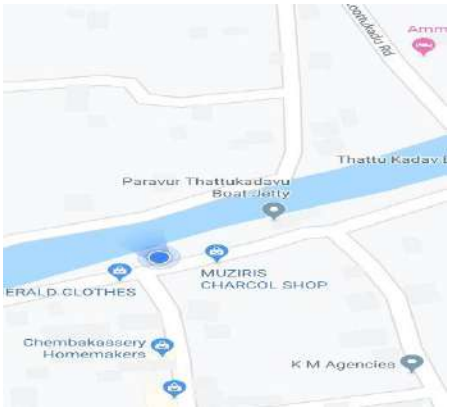

Figure 26: Location 1 Thattukadavu River

Figure 27: Sample Slide 1

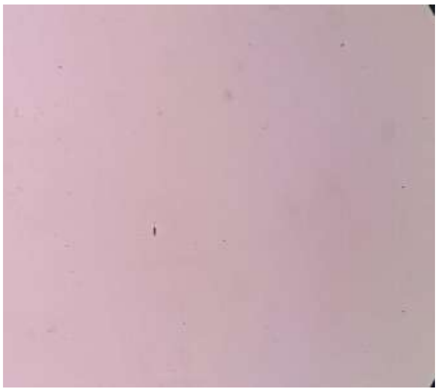

Figure 28 : Sample 1 Centric Diatoms

Figure 29: Sample slide 2

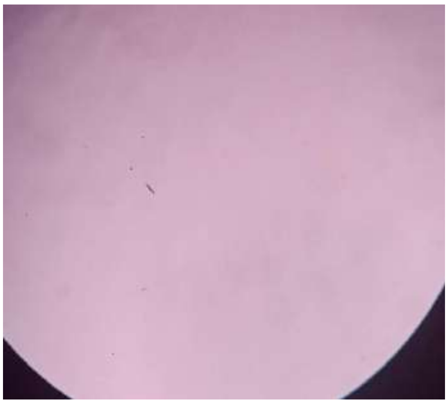

Figure 30: Sample 2 Centric Diatoms

Figure 31: Sample Slide 3

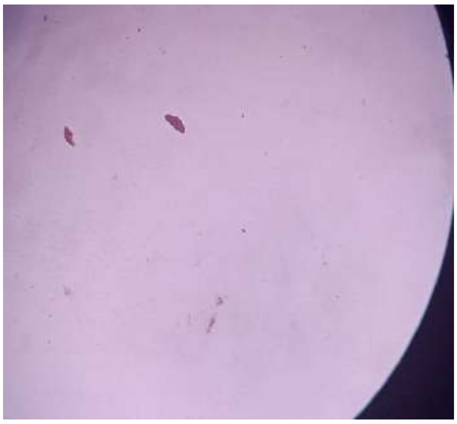

Figure 32: Sample 3 Centric Diatoms

Location 2

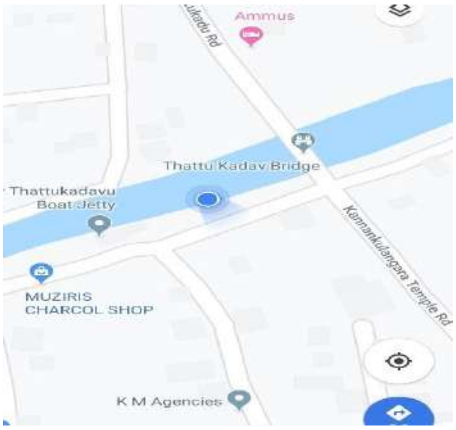

Figure 33: Location 2 Thattukadavu River

Figure 34: Sample Slide 4

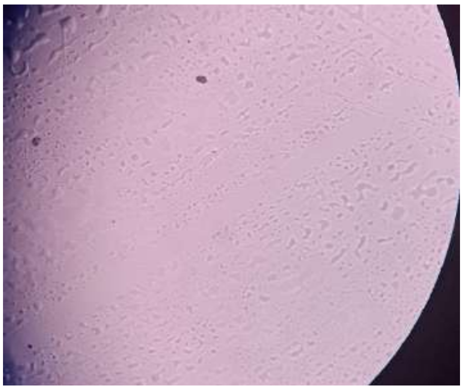

Figure 35: Sample 4 Centric Diatoms

Figure 36: Sample Slide 5

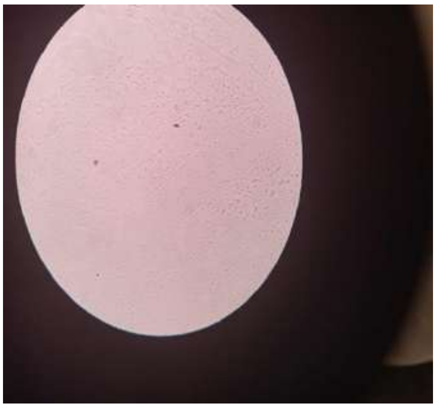

Figure 37: Sample 5 Centric and Pinnate Diatoms

Figure 38: Sample Slide 6

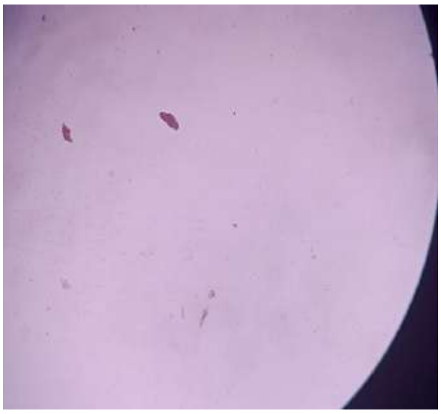

Figure 39: Sample 6 Centric and Pinnate Diatoms

Location 3

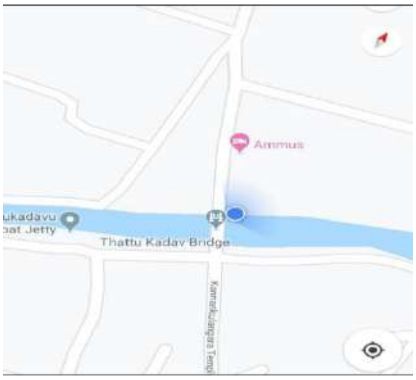

Figure 40: Location 3 Thattukadavu River

Figure 41: Sample Slide 7

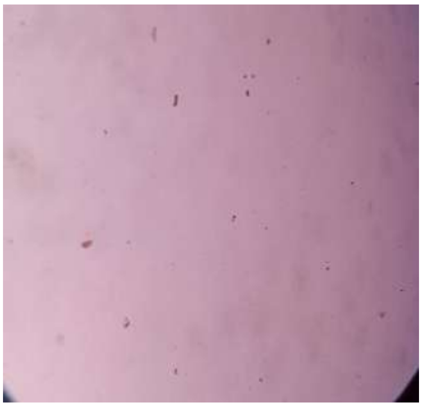

Figure 42: Sample 7 Centric Diatoms

Figure 43: Sample Slide 8

Figure 44: Sample 8 Centric and Pinnate Diatoms

Figure 45: Sample Slide 9

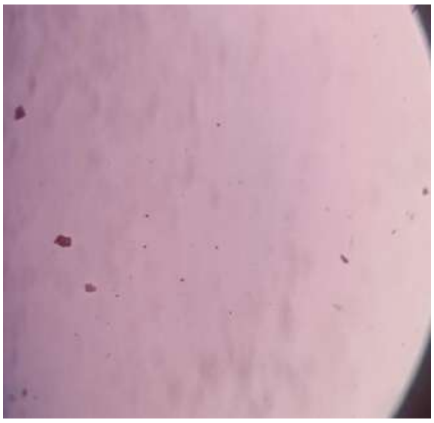

Figure 46: Sample 9 Centric and Pinnate Diatoms

Chittattukara Stream

Location 1

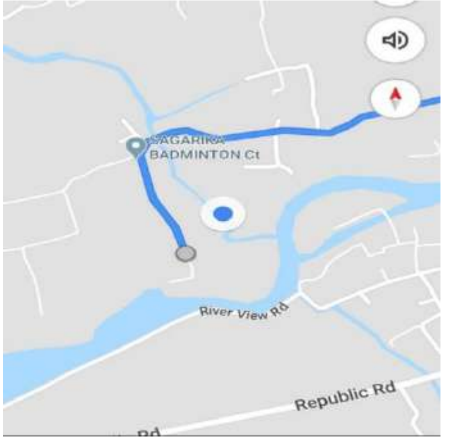

Figure 47:Location 1 Chittattukara Stream

Figure 48:Sample Slide 1

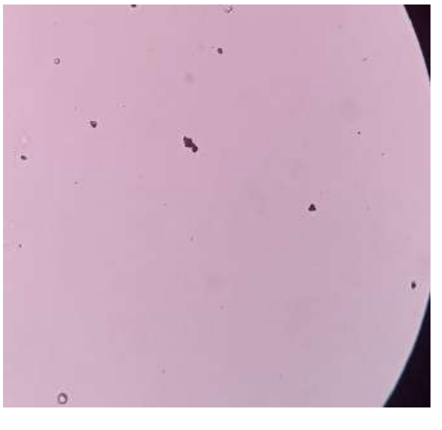

Figure 49:Sample 1 Centric Diatoms

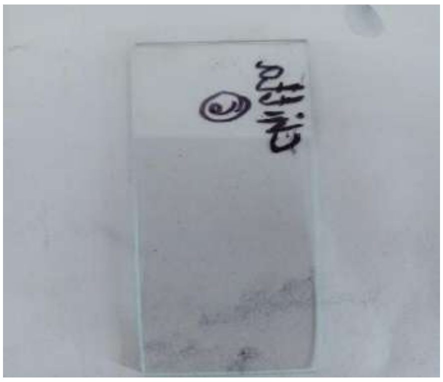

Figure 50: Sample Slide 2

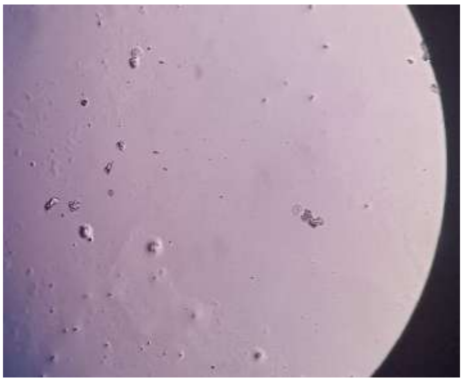

Figure 51:Sample 2 Centric Diatoms

Figure 52: Sample Slide 3

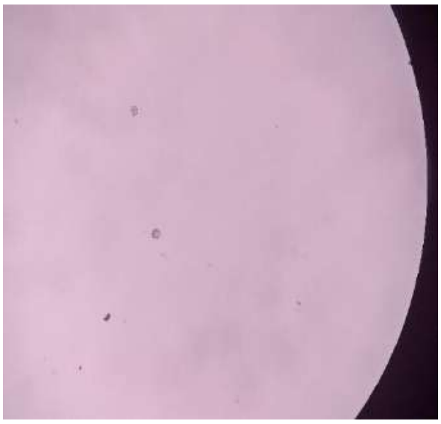

Figure 53:Sample 3 Centric Diatoms

Location 2

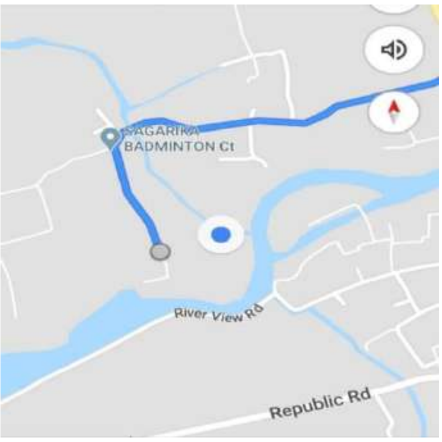

Figure 54: Location 2 Chittattukara Stream

Figure 55: Sample Slide 4

Figure 56: Sample 4 Centric Diatoms

Figure 57: Sample Slide 5

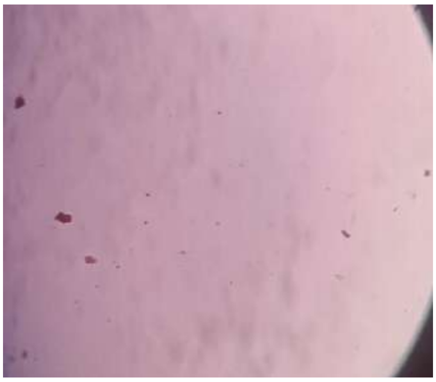

Figure 58: Sample 5 Centric and Pinnate Diatoms

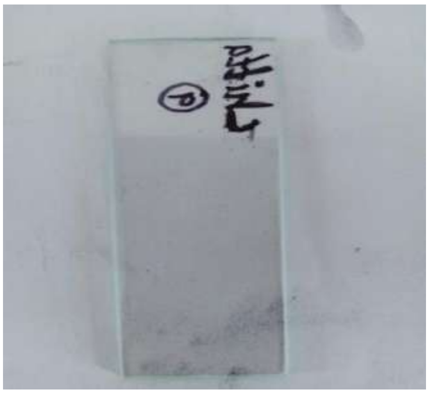

Figure 59: Sample Slide 6

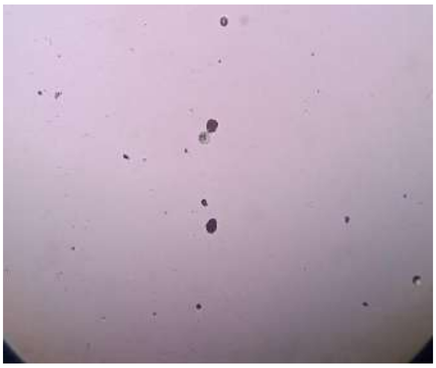

Figure 60: Sample 6 Centric Diatoms

Location 3

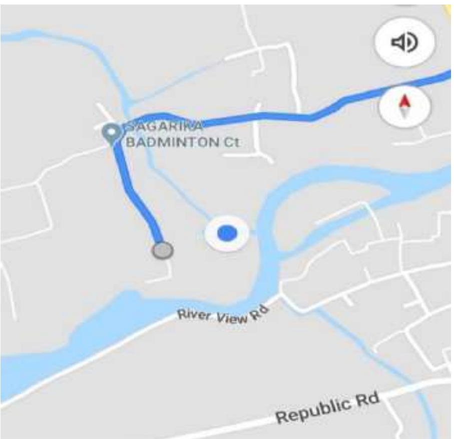

Figure 61: Location 3 Chittattukara River

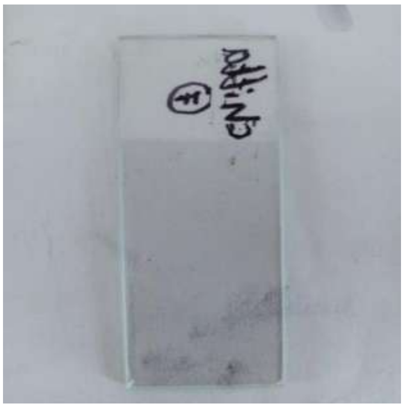

Figure 62: Sample Slide 7

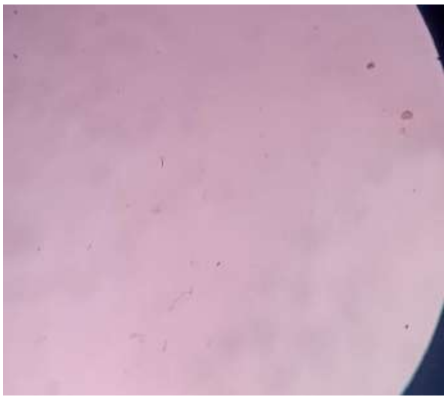

Figure 63: Sample 7 Centric and Pinnate Diatoms

Figure 64: Sample Slide 8

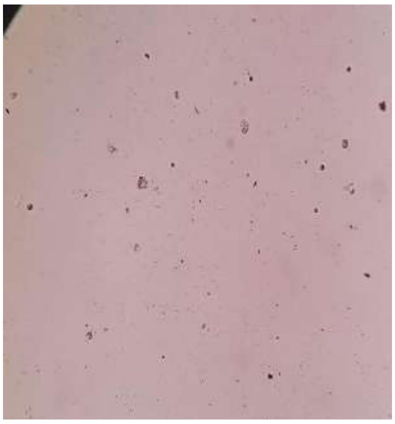

Figure 65: Sample 8 Centric Diatoms

Figure 66: Sample Slide 9

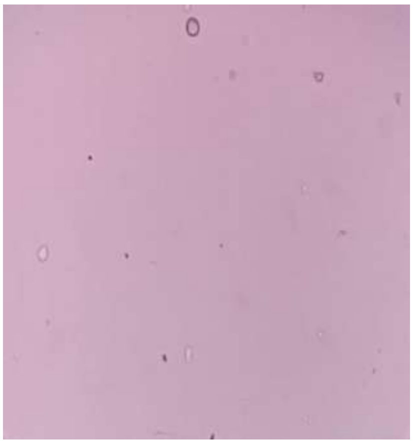

Figure 67: Sample 9 Centric Diatoms

CHAPTER VI : RESULT AND CONCLUSION

RESULT:

The two major types of Diatoms that are Pinnate and Centric Diatoms are identified in the water samples which are taken from the three locations of Chennamangalam River, Thattukadavu River and Chittattukara Stream.

CONCLUTION:

This study provides an important role for understanding the transfer of Diatoms from the water samples to the body parts. Role of Diatoms has always remained significant in solving the drowning cases. In cases, where the cause of death cannot be ascertained by conventional post-mortem examination in those cases presence of Diatoms in lungs and other body tissues plays an important role in determining whether the death is due to drowning or not.

In this study, the Diatoms present in different water bodies of North Paravoor region can be found as Pinnate or Centric. In future, study can be done in order to find the species of diatoms are present in water bodies of various regions of Kerala state.

CHAPTER VII: REFERENCES

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