"STUDY ON WATER QUALITY PARAMETERS IN VEMBAND LAKE AT MARINE DRIVE , ERNAKULAM"



Project work done by

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Affiliated to Mahatma Gandhi University, Kottayam in partial fulfillment of

requirement for the degree of Bachelor in Science in Zoology

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CERTIFICATE

This is to certify that the project entitled **"STUDY ON THE WATER QUALITY PARAMETERS IN VEMBANAD LAKE AT MARINE DRIVE, ERNAKULAM"** submitted by **Ms. ANN MARIYA**, Reg no-**AB21ZOO031** in partial fulfillment of the requirement of Bachelor of Science of science in Zoology to the Department of Zoology, St.Teresa's College affiliated to Mahathma Gandhi University ,Kottayam is a bonafide work under my guidance and supervision and to my her best knowledge, this is her best effort.

Ms. Akhila Anilkumar	Dr.Soja Louis
Assistant Professor	Head of the Department
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Ernakulam	Ernakulam

EXAMINERS

1.

2.

DECLARATION

I hereby declare that project work titled **"STUDY ON THE WATER QUALITY PARAMETERS IN VEMBANADU LAKE AT MARINE DRIVE ,ERNAKULAM"** submitted to St. Teresa's College (Autonomous), Ernakulam affiliated to Mahatma Gandhi University, Kottayam in the partial fulfillment of the requirements of Bachelor of Science degree in Zoology, is a record of original project work done by me under the guidance and supervision of Ms. Akhila Anil kumar, Assistant Professor, Department of Zoology, St. Teresa's College (Autonomous), Ernakulam.

Name: ANN MARIYA

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Signature

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ABSTRACT

One of the primary causes of the dangerously high rate of anthropogenic activities is still the low quality of the water in rivers, lakes, etc. The industrial pollutants, eutrophication, and home garbage all put a lot of strain on the Vembanad Lake.

The objective of the study was to analyse the water quality parameters and the presence of lactose fermenting bacteria like E.coli in the water collected from Marine drive area in Ernakulam of Vembanad lake. The initial sampling was done on 13 th February , total of three bottles of water samples from the site marine drive were taken. The second sampling which was the final one was taken on 20 th February that contained 3 bottles of samples taken from the same site as that of the first one.duration of the study was 1 week. The methodology involved the sample collected from the region and measuring temperature , sechi depth and colour using sechi disk, total dissolved solvent , nitrate ,BOD(Biological oxygen demand),primary productivity, standard plate count , MPN test , completed test of E.coli of the water samples taken during two different time.

The average temperature of the study is 32 °C. The sechi color which indicates a greenish brown color to the water is due to untreated effluent discharge, addition of chemical pollutants from agricultural waste, algal blooms and other pollutants. MPN test indicated that in all of the 9 tubes inoculated there is lactose fermenting bacteria. There was an average of 28mg/L of biological oxygen content in the water which indicates the water is suitable for the growth of aquatic organisms and the presence of planktons in the water source. The nitrate amount was very low in the water sample .Fluctuations in sechi depth , BOD, total dissolved oxygen , MPN were observed due to inundation , variation in daily tides , algal bloom , eutrophication etc. The study didn't have relevant values in primary productivity as it was -10 in the sample collected from sampling station.

The result embodies the need for monitoring the lake system to preserve the water quality and for assessing the level of pollution. The main of objective of the study was to analyse the physical, chemical, biological parameters of vembanad lake at specific location. The study is highly relevant since vembanad kol wetland is known for its ecological significance. It is home to more than 20,000 waterfowls – the third largest such population in India.vembanad lake was included in the list of wetlands of international importance, as defined by the Ramsar Convention for the conservation and sustainable utilization of wetlands.

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INTRODUCTION

One precious and limited resource that is essential to human survival is water. The physical characteristics of the regions where rivers flow is altered by human activity, leading to issues with water contamination. It is useful for a wide range of purposes, including drinking, irrigation, fishing, recreation, navigation, and beauty. Among the most precious natural resources is water. It is the cornerstone of the social and economic infrastructure and is necessary for both sustainable growth and a thriving community. Because of increased industrialization and population growth, water, the matrix of life, is subjected to pollution and unhealthful environments, which cause human suffering and the spread of diseases. Natural processes that support native fish populations, flora, wetlands, and wildlife are maintained by high water quality.

Due to the abundance of rainforests, Kerala, the southernmost state of India, which is located in a tropical area and on the banks of the Arabian Sea, has a humid tropical wet climate. Waters contaminated by sewage contain solids and dissolved organic compounds that give off an unpleasant smell and provide a perfect environment for microorganisms to grow and multiply. 99.9 percent of sewage is made up of water, with the remaining 0.02-0.03 percent being suspended solids and other soluble organic and inorganic materials. Even though the percentage of solids may seem low, a major municipal plant handles a massive volume of material every day that includes roughly 100 tons of solids.

Globally, the quality of the water has been connected to health outcomes. The term "drinking water quality" refers to the relationship between the natural processes and human activities that affect water composition. Water's quality is defined by its physical, chemical, biological, and aesthetic characteristics, which also determine its suitability for various applications such as safeguarding human health and the aquatic ecosystem. The majority of these characteristics are determined by substances that are suspended or dissolved in water, and both natural and man-made processes can have an impact on the quality of the water. The ability of a population to protect long-term access to sufficient amounts and suitable quality of water for sustaining.

Even though there are many places in the world where people can obtain water, it is rarely safe to drink and is rarely available in enough quantities to meet basic health needs. Open field dumping, animal waste, commercial, industrial, and agricultural operations, household waste, and animal waste are all potential sources of contaminated drinking water. fooding. This kind of contamination can affect any source of water, in particular. Sanitation and access to clean drinking water are major global issues.

Punnamada, commonly known as Vembanad Lake, is one of the longest and largest lakes in India. It is thought to be among Kerala's most picturesque locations. It is located on the right side of the Arabian Sea, separated from the ocean by a short reef. This lake is also the nation's second-largest Ramsar reserve, after the Sunderbans in West Bengal.

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The Vembanad Lake in India map depicts the river's location in Kerala. The lake is 96.5 kilometers long, 14 kilometers wide, and 12 meters, or 39 feet, deep. The National Wetlands Conservation Programme has also acknowledged it. Vembanad-Kol Wetland is the lake's official name. The Ramsar Convention lists this lake as one of the wetlands of international concern. For this reason, the convention strives to maintain and protect this wetland. In addition, it provides over 20,000 ducks with refuge and a suitable environment for shrimp. The lake's resources of clams and subfossils are also noteworthy. The lake provides several benefits to nature. It facilitates streamflow management, drought mitigation, and groundwater aquifer replenishment. This lake serves as a habitat for both aquatic and semi-aquatic plants and animals, which further supports the survival of terrestrial species. In addition, a large number of residents rely on the lake. They work in agriculture, inland navigation, coir retting, fishing, tourism, and lime shell production.

Vembanad Lake is home to a lot of kinds of fish and birds, and even has a bird sanctuary closeby. It also provides sustenance to 16 million people by providing water as well as means of livelihood in the form of fishing, tourism, agriculture, etc. Vembanad Lake is also a National Waterway because of its well connectedness via canals. Its water flows out through different canals which flow 196 kilometres from north to south and 29 kilometres from east to west. In this way, the lake is also very well connected to a lot of villages surrounding Kottayam, Ernakulam and Alleppey.

In terms of ecology, the Vembanad Lake is incredibly important as well. It has the third largest population of waterfowls in India, is the perfect home for various fish, and is even home to over a hundred breeds of birds. In fact, the Kumarakom Bird Sanctuary is close to the lake as well. The sources of Vembanad Lake are a few freshwater rivers, namely Achankovil, Manimala, Pamba, Periyar, Muvattupuzha and Meenachil Rivers. The lake outflows through its canals which spread for vast distances in all four directions. It also encompasses several islands, such as Perumbalam, Pathiramanal, Palippuram, Willingdon Island and Vallarpadam island.

The 1,252 m (4,108 ft) Thanneermukkom salt water barrier, which was built as part of the Kuttanad Development Scheme to stop tidal action and salt water intrusion into the Kuttanad lowlands, is one of the lake's distinctive features. It effectively splits the lake into two sections, one with fresh water from rivers that drain into it and the other with permanent brackish water. It is the largest mud regulator in India. Because it removes salinity from the area and gives farmers in Kuttanad an extra crop to grow during the dry season, this barrier has benefited them. One of the Vembanad Lake's narrower points is where the Thanneermukkom barrier is situated. In July, only two-thirds of the gates that were initially opened are opened to release floodwaters.

The structure's primary disadvantage has been the loss of fish and prawn migration opportunities upstream, as well as an increase in weed growth in the upstream, which has significantly limited the natural flushing of pollutants. The Thanneermukkom bund has also led to ecological issues, chief among them being the Water Hyacinth's unchecked spread throughout freshwater environments.

Three islands, Pathiaramanal, Pallippuram, and Perubalam, encircle the lake and span an area of more than 2033.02 km2. Ten rivers, including Kerala's six principal rivers, feed the lake. The total area that the lake drains is approximately 15,770 square kilometers.

However, like all other water bodies in India, even Vembanad Lake is highly polluted with several pollutants, such as microplastics, plastic, metal, glass, cloth, sewage, pesticides, etc. This is proving to be severely harmful for the aquatic life living in the lake.

The lake's water spread area has been steadily declining over time. For example, the area was 290.85 square kilometers in 1917, 213.28 square kilometers in 1971, and 213.28 square kilometers in 1990. According to research, land reclamation has caused the area to shrink by around 37% of its original size, posing a serious ecological concern. Pesticides and heavy metals have severely contaminated this lake. Residents near the lake now face health hazards as a result of this. Research has demonstrated that contamination can cause the biological body's biochemical processes to become

Furthermore, the zone in Kerala has been significantly affected by industrial activities and household waste. As a result, the Vembanad estuary's depth and transparency have decreased, and its dissolved oxygen level has also decreased. The primary causes of the drop in oxygen levels are sewage discharge and houseboat tourism. Significant contamination has resulted from development activities, waste dumping, and rusted boats in the Alappuzha canals.

Objective :

- To study the water quality parameters in vembanad lake .
- The parameters taken to check the water quality of water are of three categories. That are physical, chemical and biological parameters .Physical parameters include Temperature, colour, sechi depth and TSS. Chemical parameters are nitrate, BOD and primary productivity. Biological parameters are standard plate count, MPN test and completed test of E.coli. The sampling was done two times with one week apart.
- Analyzing the quality of water contributes to the identification of possible health hazards linked to tainted water sources. This involves keeping an eye out for microbiological pollutants including bacteria, viruses, and parasites that can lead to gastroenteritis, cholera, and other waterborne illnesses..
- Data for risk assessment and management plans are provided by the analysis. Authorities can put policies in place to reduce pollution and protect water resources by determining possible threats and their sources..
- Sharing the results of the study with the public raises awareness about water-related issues and encourages community participation in water conservation and pollution prevention efforts.

LOCATION TAKEN FOR SAMPLING

Marine Drive is one of the popular hangouts in Ernakulam city. Facing the serene backwaters with a walkway of about three kilometers, the place attracts a lot of local residents as well as tourists every day. The backdrop of the backwaters, with ships anchored at the harbour, is one of the main reasons for its allure.

The walkway of Marine Drive begins from the High Court Junction and continues up to Rajendra Maidan. You will also find boat jetties here that add a distinct charm to the whole place. Marine Drive is also known as the Queen's Necklace because, when viewed at night from an elevated point anywhere along the drive, the street lights resemble a string of pearls in a necklace. Latitude and longitude of the sampling station is (9.9785°N 76.2763°E). First sampling was done on 13th February 2024 and the second sampling was done



Image1: of Marine drive the sampling station for water collection

REVIEW OF LITERATURE

The study done on the topic Drinking water quality assessment and its effects on residents health in Wondo genet campus, Ethiopia by Yirdaw Meride* and Bamlaku Ayenew(2016) found that the drinking water at Wondo Genet Campus was in good condition, with a mean turbidity of 0.98 NTU and an average temperature of 28.49°C. The water had a high concentration of dissolved solids, chloride, sulfate, magnesium, calcium, sodium, and potassium. The water was also analyzed for total coliform bacteria, indicating that it met the World Health Organization's standards for drinking water.

The study on Water quality assessment and evaluation of human health risk of drinking water from source to point of use at Thulamela municipality, Limpopo Province done by N. Luvhimbi *et al.* (2022)was evaluated the physicochemical and bacteriological quality of drinking water in Thulamela municipality, Limpopo Province, South Africa. The water had a mean turbidity of 0.98 NTU, average temperature of 28.49°C, and a mean total dissolved solids concentration of 118.19 mg/l. The water had a chloride mean value of 53.7 mg/l and a sulfate mean value of 0.33 mg/l. The water had a magnesium concentration of 13.67 mg/l, calcium concentration of 2.16–7.31 mg/l, sodium of 31.23 mg/l, and potassium of 23.14 mg/l. The water samples were analyzed for total coliform bacteria and found to be consistent with the World Health Organization standard for drinking water. The study recommends a more consistent supply of treated municipal water and training residents on hygienic practices for transportation and storage of drinking water.

A water quality assessment was conducted in Timchirappalli, Tamilnadu, India, done by G. Venkatesan *et al.*,(2013) to assess the potential pollution of water resources due to distillery industry effluents. Samples were collected from various locations around the distillery unit and analyzed for chemical parameters like hardness, pH, chlorides, sulphates, alkalinity, total dissolved solids, nickel, chromium, dissolved oxygen, biological oxygen demand, and chemical oxygen demand. Surface water samples were analyzed for parameters like BOD, COD, TDS, copper, zinc, lead, cadmium, nickel, and manganese. The parameters were checked against World Health Organization standards, and it was found that some parameters, such as hardness, were 10 times higher than pure drinking water.

The investigation done on Microbiological and Physicochemical Water Quality Assessments of River Water in an Industrial Region of the Northwest Coast of Borneo by Sui S. Leong *et al.* (2018) the water quality (WQI) of river water in an industrial region northwest of Borneo, focusing on physicochemical and microbial characteristics. The microbiological parameters tested included total viable count (TVC), coliform count (TC), faecal coliform count (FC), and Escherichia coli confirmation. Physicochemical constituents evaluated included water temperatures, pH, total dissolved solids (TDS), salinity, electrical conductivity (EC), dissolved oxygen (DO), biochemical oxygen demand (BOD), ammoniacal nitrogen (NH3-N), chemical oxygen demand (COD), and total suspended solids (TSS). The mean microbial counts of samples were significantly different

between the rivers, with the tested parameters exceeding international limits. The WQI ranged from 65-73 under class III, with most water being slightly polluted and posing a potential public health threat.

A study in Nigeria done by Samuel O. Olasoji *et.al.*(2019) evaluated the quality of 12 water sources and 2 treated water used by a peri-urban town to assess their suitability for drinking and domestic use. The water quality parameters included pH, temperature, acidity, total alkalinity, chloride content, and total CO2. The study used a Flame Atomic Absorption spectrophotometer to determine the concentrations of Ca, Mg, Cu, Cr, and Pb in the samples. The majority of the water samples (86%) were rated as excellent, with one sample each rated as poor and good water quality. The study recommends including microbial water quality parameters in all Water Quality Index (WQI) analyses to provide a true status of a water resource's quality.

Ranjith Kumar .P *et.al.*,(2017) conducted a study on the topic Physico-chemical characteristics of Vembanad backwaters at Eramalloor region, Alappuzha district, Kerala, India.The study was conducted on Vembanad lake in Kerala from September 2015 to July 2016, observing average temperatures of 29.0°C, salinity of 19.02%, transparency of 71.60 cm, lowest dissolved oxygen in March, maximum pH in May, and minimum in October. The average alkalinity was 90.98 mg/l, with nitrate and phosphate levels ranging from $16.3\mu g/l$ to $5.8\mu g/l$. The peak salinity was observed during the premonsoon season, and mesohaline nature was observed throughout the study period.

S.Sruthy ,E.V Ramaswamy(2017) presented the study on Microplastic pollution in vembanad lake,kerala, India. The study presents the first study on microplastics in the sediments of Vembanad Lake, a Ramsar site in India. Microplastics are emerging pollutants with a particle size of <5 mm, originating from the degradation of larger plastic debris or manufactured as small granules. The impact of microplastic pollution on the environment and biota is not well known, and data on freshwater ecosystems is scarce. Samples were collected from arine microplastics and processed for microplastic extraction through density separation. Micro Raman spectroscopy was used to identify the polymer components of MPs. MPs were found in all sediment samples, indicating their extensive distribution in the lake. The abundance of MPs recorded ranged from 96-496 particles m, with low density polyethylene being the dominant polymer component. The presence of MPs in the lake poses a severe threat to the local population's food web. This study is the first from India on MPs in lake sediments, urging further research on the distribution and impact of emerging pollutants on aquatic systems across India.

Meera s and Bijoy nandan(2010) studied _Water quality status and primary productivity of Valanthakad Backwater in Kerala .The study monitored the water quality and primary productivity of Valanthakad backwater from June to November 2007. It found significant variations in temperature, transparency, salinity, pH, dissolved oxygen, sulphides, carbon dioxide, alkalinity, biochemical oxygen demand, phosphate-phosphorus, nitrate-nitrogen, nitrite-nitrogen, and primary productivity. Transparency was low during the active monsoon months, resulting in turbid waters. Salinity was low except in August and November 2007. Total sulphide and higher carbon dioxide were found due to hospital discharges, decaying slaughter house wastes, and mangrove

vegetation. Nitrate-nitrogen and phosphate-phosphorus values were higher during the monsoon season. Maximum net primary production was observed in November. Chlorophyll pigments showed higher values in July, August, and November, with a negative correlation with phosphate-phosphorus and nitrite-nitrogen.

Mani, K. *et.al.*, (2013) studied Spatial Distribution of Non-Point Source Pollution of Vembanad Lake, Kerala, .This study analyzed pollution levels and spatial distribution of pollution parameters on the Vembanad lake in Kerala. The analysis involved plotting spatial and temporal variations across 35 sites and in post-monsoon and pre-monsoon seasons. The parameters analyzed included acidity, hardness, chloride, sulphate, COD, and iron. The results helped identify more polluted areas and suggest measures to improve water quality and control waste outflow into the lake.

A study in Kerala, India, done by M.Vaiyapuri *et al.*, assessed the antimicrobial resistance (AMR) burden in Escherichia coli along the 90 km stretch of Vembanad Lake(2021). The study found that 77% of water samples from 35 stations harbored E. coli. The antibiotic susceptibility test revealed resistance to at least one antibiotic in 81% of E. coli isolates, with multidrug resistance in 30% and extended spectrum β -lactamase (ESBL) producers in 32%. The highest probability of isolating cefotaxime-resistant E. coli was found in the lake. Genetically diverse ESBL types, including blaTEM-116, blaCTX-M-152, blaCTX-M-27, blaCTX-M-55, blaCTX-M-205, and blaSHV-27, were identified in the lake. The low multiple antibiotic resistance index suggests a lower risk to the human population, but the occurrence of genetically diverse ESBL E. coli in the lake signals health hazards and requires pragmatic control measures.

METHODOLOGY

PHYSICAL PARAMETERS

EXPERIMENT 1

Temperature of the respective water sample are measured using a thermometer.

EXPERIMENT 2

The Secchi depth is measured using standard protocols: The white Secchi disk is lowered into the water and the depths at which it disappears and reappears are recorded. The Secchi depth is computed by averaging these two depth measurements. It may be feasible to view the depth of disk disappearance and reappearance directly from the measuring tape at the water surface. If this is not feasible, the distance from the hand-held device (casing) to the water surface (DO) can be measured, as well as the total distance (TO) from the device to the depth at which the disk disappears and reappears. The Secchi depth can then be computed by subtracting DO from TO Note if the latter is done, it is essential to measure DO accurately and to keep DO constant throughout the measurement (eg, by maintaining arm at 90 degrees).

For accurate Secchi depth readings, the observer should avoid sun glint regions and shadows, ideally conduct the measurement closer to mid-day (or at the very least record the time and location which can be used to compute sun angle), allow their eyes to adapt near to the Secchi depth, write down sky conditions, and repeat measurements to improve precision.

The disk must sink vertically though the water for accurate Secchi depth readings. The weight of the mini-Secchi (100 g) should be sufficient for vertical deployment from a fixed platform in waters with low current speed. However, in stronger currents, or in 12 cases where the platform may be moving (e.g, from a boat) extra weight will be required to avoid the disk sinking at an angle.

The colour of the water is measured by looking at the colour of the Secchi disk at roughly half the Secchi depth, matching it to the closest colour on the colour scale and noting the corresponding number. Once the disk is at half the Secchi depth, it is relatively straight-forward to turn the hand holding the device so that your palm is facing up and the scale is visible and you can see the disk .After operating the device, the mini-Secchi should be cleaned with fresh water and stored in a dry location. To remove any dirt from the white disk a little washing liquid may be used.

EXPERIMENT 3

Total dissolved solvent

Aim: Determine the Total dissolved solids (TDS) of given water samples.

Introduction: Water is a good solvent and picks up impurities easily. Pure water tasteless, colorless, and odorless is often called the universal solvent. Dissolved solids" refer to any minerals, salts, metals, cations or anions dissolved in water. Total dissolved solids (TDS) comprise inorganic salts (principally calcium, magnesium, potassium, sodium, bicarbonates, chlorides, and sulfates) and some small amounts of organic matter that are dissolved in water. In general, the total dissolved solids concentration is the sum of the cations (positively charged) and anions (negatively charged) ions in the water. Therefore, the total dissolved solids test provides a qualitative measure of the amount of dissolved ions but does not tell us the nature or ion relationships. An elevated total dissolved solids (TDS) concentration is not a health hazard. The TDS concentration is a secondary drinking water standard and, therefore, is regulated because it is more of an aesthetic rather than a health hazard. An elevated

TDS indicates the following:

1)The concentration of the dissolved ions may cause the water to be corrosive, salty

or brackish taste, result in scale formation, and interfere and decrease efficiency of

hot water heaters; and

2)Many contain elevated levels of ions that are above the Primary or Secondary

Drinking Water Standards, such as an elevated level of nitrate, arsenic, aluminum,

copper, lead, etc.

Requirements:

Water sample to be tested

Evaporating Dish/ Ceramic Dish

Desiccator

Whatman Filter paper

Electric balance machine

Procedure:

1. Filter your water sample through a Whatman Filter paper.

2. Collect the filtrate (liquid) and rinse water in a flask.

3. Take the weight of empty container (ceramic dish/ evaporating Dish). Make sure the container should be dried.

4. Add the filtrate to the container and allow the sample to stay in the oven at 103°C for 24 hours. If possible, increase the temperature of the drying oven to 180°C and allow the sample to dry for up to 8 hours.

5. Remove the container - Remember it is very hot. After removing from the drying oven, the sample should be placed in a desiccator to cool in a dry air environment for at least 3 to 4 hours.

6. After the container cools, reweigh the container at least three times.

7. Subtract the initial weight (in grams) of the empty container from the weight

of the container with the dried residue to obtain the increase in weight. Then

do the following:

A- Weight of clean dried container (gm)

B- Weight of container and residue(gm)

C- Volume of Sample (ml)

Concentration (mg/L) = ((B - A)/C)* (1000 mg/g) * (1000 ml/L)

For example:

A=100.0001gm

B=100.0020gm

C=100ml

Concentration (mg/L) = ((100.0220 - 100.0001)/ 100) * 1000 * 1000 = 219 mg/L

Chemical parameters

EXPERIMENT 4

Estimation of nitrate

In terrestrial and aquatic ecosystem, nitrogen occurs in various forms such nitrogen gas(N2) compounds like ammonia (NH3), inorganic ions like nitrate (NO3) and nitrite (NO₂), organic molecules like proteins, peptides, etc. Many of them are part of nitrogen cycle. Nitrate are essential plant nutrients. Nitrates generally occur in trace quantities in surface water but may attain high level in some ground water. Nitrate values are commonly reported as either nitrate on as nitrate - nitrogen. The maximum contaminant level in drinking water as nitrate (NO3) is 45mg/L, whereas the MCL is 10mg/L 2 No3-N Nitrate is usually very little in fresh domestic waste water but may be very high in some industrial effluents. The source of excess nitrates, can usually be traced to agricultural activities such use of nitrogenous fertilizers, human and animal pollution high concentration of nitrates in water sources may cause many deleterious effects in human beings and animals.

For determination of nitrates (Noz) usually complex procedures are in use. This is a simple method for the determination of 'nitrate' content of water samples based on the rapid colorimetric method given by cataldo et.al (1975).

PRINCIPLE

The complex formed by the nitration of salicylic acid under highly acidic condition absorb maximally 410nm in a basic solution. Absorbance is directly proportional to the amount of nitrate - nitrogen present. Ammonium, nitrate and chloride ions do not interfere.

REAGENTS:

1)Salicylic acid - H2SO4 reagent

5% of Salicylic acid in conc. H2SO4 (Weight / volume) Dissolve 5g of Salicylic acid in 100ml of conc. H2SO4. reagent should be made fresh every week and stored in a brown bottle).

2)2N NaOH

In a 250ml beaker, take 100ml of water and dissolve 40.0g of NaOH pellets. Transfer it to a 500ml volumetric flask and make upto 500ml with distilled water.

PREPARATION OF STANDARDS

Stock solution - 0.25g/L NO3-N(= 250mg/L or 250 mg/ml): Dissolve 1.805g potassium nitrate (kno3) approximately 600 ml disttiled water taken in a 1L standard flask. Ensure all kno3, is dissolved, make upto the mark with distilled water mix well and store in a at 4°c suitably labelled container.

Blank: A blank of 0.025 ml distilled water with all the reagents is normally sufficient .if the sample is pigmented a separate blank may be required .Prepare the blank with 0.25 ml sample ,0.8 ml conc. Sulphuric acid and 19 ml of 2N NaOH.

Procedure

1. Arrange 3 test tubes marked 'Test', 'standard' and Blank on a test tube rack

2. Pipette 0.25 ml sample to the test tube marked "Test". 0.25 ml nitrate standard solution to the test tube marked standard' and 0.25ml distilled water to the test tube marked 'Blank'.

3. Add 0.8ml of 5% Salicylic acid in conc. H2S04 to all the 3 test tubes and miX thoroughly (the wine red Colour develops in Test tube and standard' will turn golden yellow after adding NaOH. The white precipitate formed in the blank after adding NaOH). the Blank will completely dissolve after adding NAOH

4.After 20 minutes add 19ml of 2N NaOH to raise the PH above 12

5.Cool samples to room temperature

6.Measure absorbance at 410mm with a Spectrophotometer

7.Calculate the concentration of the Sample using the formula. (concentration of the sample can diluted using a also be standard curve prepared with a Series of different concentrations of KNO3)

EXPERIMENT 5

DISSOLVED OXYGEN

AIM : To estimate the amount of oxygen dissolved in water using Winkler's method .

PRINCIPLE : In Winkler's method , nascent oxygen (O) released from water is used to liberate iodine from potassium iodide and this iodine is estimated using standard sodium thiosulphate (Na2S2O3) . When manganese sulphate (MnSO4) is added to the sample of water followed by strong alkaline potassium iodide , manganese hydroxide (Mn(OH)2) will be formed. This combines with dissolved oxygen in water to form manganic hydroxide (Mn(OH)3 . This on acidification with concentrated sulphuric acid in the presence of alkaline potassium iodide 23 releases iodine equivalent to the amount of oxygen used in this reaction. The amount of iodine liberated is estimated by titrating the sample solution against 0.01N sodium thiosulphate.

 $MnSO4 + 2KOH \rightarrow Mn(OH)2 + K2SO4$

$2Mn(OH) \ 2 + H2O + \frac{1}{2}O2 \rightarrow 2 \ Mn(OH)_3$

$2 \ Mn(OH)_3 + 2KI + 3H2 \ SO4 \rightarrow 2Mn \ SO4 + K2SO4 + 6 \ H2O + I2$

$2Na2S2O3{+}I2 \rightarrow Na2S4O6{+}2NaI$

 ${\bf REQUIREMENTS}$: Manganese sulphate , alkaline potassium iodide, conc. sulphuric acid , sodium thiosulphate , fresh starch solution (as indicator) , burette , pipette , conical flasks , measuring cylinders , beakers , reagent bottles , oxygen estimation bottles with stopper and samples of water .

1. Manganese sulphate solution : (480 gm MnSO4 . 4H2O or 400gm MnSO4 . 2 H2O or 364gm MnSO4 . H2O in 1 litre distilled water)

2. Alkaline iodide solution : (KOH - 700gm and KI - 150gm in 1 litre distilled water)

3. Sodium thiosulphate solution - 0.1N Na2S2O3 (24.82gm Na2S2O3 . 5 H2O in 1 litre distilled water) Dilute this stock solution 10 times (1ml to 10 ml) to get 0.01 N solution .

PROCEDURE 1. FIXATION OF WATER SAMPLES

Water sample is taken in a bucket and a 250 ml reagent bottle is immersed in it with minimum disturbance. Taking care to avoid any air bubbles inside it, close the bottle with the stopper under water. Take out the bottle and remove the stopper. To the water sample in the bottle add 1 ml of manganese sulphate solution followed by 1 ml alkaline potassium iodide solution . Add the reagents well below the surface by keeping the tips of the pipette near the bottom and gradually pulling them upwards. Close the bottle tightly with the stopper and avoid air bubbles inside. Shake the sample very well in order to complete precipitation inside. Leave the mixture undisturbed for sometime so that the precipitate settles down completely.

2. TITRATION OF WATER SAMPLE

Add 2 ml of conc. H2SO4 carefully to the precipitate. Close the bottle with the stopper and mix the contents very well to dissolve the precipitate completely . From the straw coloured solution thus formed, pipette out 20 mlinto a clean conical flask . Titrate against 0.01 N sodium thiosulphate solution from the burette . When the solution turns pale yellow add a few drops of freshly prepared starch solution as indicator. Continue the titration till 24 the disappearance of the blue colour. Note down the burette reading and repeat the titration for concordant values.

EXPERIMENT 6

PRIMARY PRODUCTIVITY

AIM: Estimation of Primary Productivity in freshwater bodies

Introduction: The primary production in the aquatic ecosystem starts with the synthesis of organic compounds from the inorganic constituents of water by the activity of plants / phytoplankton in

the presence of sunlight. The inorganic constituents which form the raw material for this synthesis are water, carbon dioxide, nitrate ions, phosphate ions and various other chemical substances. The products are mainly carbohydrates and proteins and fats in very small quantities. Organic production by plants is the first step in tapping energy by living beings from non-living natural resources and hence called primary productivity.

The method of estimating primary productivity by dark and light bottle method was introduced by Garder and Gran (1930). In this method, the water samples are incubated for a certain period in light and dark bottles which are then suspended at the same depths from where the samples are taken. In light bottles, oxygen is released as a result of photosynthesis and a part of oxygen is used for community respiration. In the dark bottles, only oxygen consumption takes place as a result of respiration. The amount of oxygen liberated by phytoplankton during photosynthesis is considered as a measure of primary production.

Material required

BOD bottle (2 light / transparent and 1 dark), Nylon or Jute ropes, Burettes, Reagents (Manganese sulphate solution, Alkaline iodide solution, Sodium thiosulphate, Concentrated Sulphuric acid, Starch indicator solution) etc.

Procedure:

a. Fill three BOD bottles with water sample in round stoppered bottles (1 Light bottle, 1 dark bottle and 1 control light bottle) avoiding air bubbles.

b. Water sample in the control bottle is immediately fixed by using Winker's fixatives

c. The dark bottle is wrapped with aluminum foil and kept in a black bag to protect from light.

d. Use one of the light bottles for estimating the initial dissolved oxygen As control)

e. Suspend both light and dark bottles exactly at the depth from where the sample was drawn are then suspended on to a raft and anchored.

f. The bottles are normally incubated for a period of 3-4 hrs between dawn to midday or sunset in the respective depths.

g. At the end of incubation period, the bottles are retrieved and fixed with oxygen fixatives. h. The oxygen content in the sample is determined by using Winkler's method.

Calculation

Let the initial oxygen level be- IB

Let the final oxygen level in dark bottle be - DB

Let the final oxygen level in light bottle be - LB Net oxygen production - LB – IB Oxygen consumed for respiration - IB – DB Gross production of oxygen - LB – DB Let 't' be the number of hours of incubation Therefore the Primary productivity can be calculated from the formula: **Gross primary productivity** = LB – DB x 1000 x 0.375 /1.25 x t mg C/m3 /hour **Net primary productivity** = LB = IB x 1000 x 0375/1.25 x t mg C/m3 /hour **Community respiration rate** = IB – DB x 1000 x 1 x 0.375 / 1.25 x t mg C/m3 /hour

Biological parameters

EXPERIMENT 7

PRESUMPTIVE TEST

Enumeration of total coliform The MPN method is a well-established and fully documented method of estimating the number of viable microorganisms in environments with extremely low level of health indicators. It estimate a microbial population size based on a process-related attribute. Common examples include growth, enzyme action, or catalytic chemistry. The MPN method involves taking the original solution or sample, and subdividing it by orders of magnitude (frequently 10x or 2x), and assessing presence/absence in multiple subdivisions. The degree of dilution at which absence begins to appear indicates that the items have been diluted so much that there are many subsamples in which none appear. A suite of replicates at any given concentration allow finer resolution, to use the number of positive and negative samples to estimate the original concentration within the appropriate order of magnitude. The samples are incubated and mostly assessed by eye or colour detection (use of specific reagents). The major weakness of MPN methods is the need for large numbers of replicates at the appropriate dilution to narrow the confidence intervals.

Enumeration of total coliform count

The coliform group comprise all aerobic and facultative anaerobic Gram negative non spore forming rod shaped bacteria that ferment lactose with gas and acid formation within 48h at 37 °C. The coliform examination is performed by adding measured volumes of sample to multiple

dilution tubes containing appropriate medium and incubating them for 48h at 37 °C, and the coliform counts are computed from number of tubes showing gas and acid production. Durham's tubes are inserted into each tube to monitor gas production. Brilliant green lactose broth (BGLB is confirmative test for coliform. Bile salts and Brilliant green almost completely inhibit the growth of other lactose fermenting bacteria like clostridium.

Materials Required

Lactose broth (HiMedia)

Brilliant Green Lactose Bile Broth

McCartney tube

Pipettes

Durham's tube

Procedure

1. Prepared 30 ml of Lactose broth in double strength (double strength is prepared so as to reduce the effect of dilution that occurs on addition of large volume of sample) and 60 ml in single strength

2. Dispensed the double strength medium into three tubes (10 ml in each tube) and single strength into 6 tubes (10 ml in each tube)

3. Durham tube was inserted into each tube, care was taken to avoid air bubbles

4. Autoclaved at 15lbs pressure for 15min

5. Inoculated 10 ml sample into double strength LC broth

6. Inoculated 3 tubes of single strength with 1 ml and other 3 tubes with 0.1 ml sample

7. Incubated all tubes at 37 °C for 48 h and document turbidity and gas production

8. MPN values were computed from the number of positive tubes.

EXPERIMENT 8

MPN - Completed test for *E. coli*.

To perform the completed test for *E. coli*, gently agitate each gassing EC tube, remove a loopful of broth and streak for isolation on a MacConky agar plate and incubate for 18-24 h at $35^{\circ}C \pm 0.5^{\circ}C$.

Streak a plate of MacConkey's agar with the desired pure culture or mixed culture. If using a mixed culture use a streak plate or spread plate to achieve colony isolation. Good colony separation will ensure the best differentiation of lactose fermenting and non-fermenting colonies of bacteria.E.coli microorganisms grow on this selective media because they are gram-negative non-fastidious rods. Growth of E. coli, which ferments lactose, appears red/pink on the agar.

EXPERIMENT 9

Standard plate count

Principle

The Plate count method is based on the idea that each viable bacterium will proliferate and form a distinct colony when an agar media containing microorganisms is incubated. This procedure involves properly mixing a particular volume, often 1 mL, of the serially diluted liquid sample with around 15 mL of molten agar medium at a temperature of 40–45°C (less than 50°C) in a Petri plate.

REQUIREMENTS

- 1. **Sterilization equipment**: Autoclave or sterilizer to sterilize all the equipment, materials, and media before use.
- 2. **Dilution blanks**: Glass test tubes or sterile pipette tips for preparing serial dilutions of the sample.
- 3. **Plate count agar**: Petri dishes containing agar medium for culturing microorganisms. The type of agar used will depend on the microorganisms being tested.
- 4. **Inoculating loops**: Metal loops are used to transfer the diluted sample onto the agar plates.
- 5. Colony counter: A device used to count the colonies that form on the agar plates.
- 6. **Incubator**: A device used to provide optimal conditions for the growth of microorganisms, including temperature and humidity control.
- 7. **Pipettes**: Single-channel or multi-channel pipettes for dispensing precise volumes of liquids during the dilution process.
- 8. **pH meter**: A device used to measure the pH of the media to ensure that it is within the optimal range for microbial growth.
- 9. **Sterile water**: Water that has been sterilized and used for diluting the sample and preparing the media.
- 10. **Sterile media**: Liquid or solid media used for culturing microorganisms. The type of media used will depend on the microorganisms being tested.

PROCEDURE

- Write 10^{-1} , 10^{-2} , 10^{-3} , 10^{-4} , 10^{-5} in the dilution tubes.
- Prepare the first dilution by putting 1 ml or 1 g of the sample into a 9 ml dilution blank marked 10-1. This dilutes the original sample 10 times
- To make sure the organisms are spread out evenly, mix them well (cells).

- From the first dilution, move 1 ml of the suspension while in motion to the dilution blank 10⁻² with a clean and sterile 1 ml pipette. This will dilute the original specimen/suspension 100 times (1/100 or 10⁻²).
- With a new, clean pipette, move 1 ml of the 10⁻² suspension to the 10⁻³ dilution blank. This makes the original sample 1000 times weaker (10⁻³).
- Use a new, clean, sterile pipette each time until the original sample has been diluted 10⁻⁵ times.
- With the right pipettes, add 1ml or 0.1ml of suspension from the right dilutions (10⁻¹ to 10⁻⁷) to sterile Petri dishes while moving. For each dilution, you need to use 2 or 3 Petri dishes.
- Melt the nutrient agar medium and let it cool. Add about 15 ml of the melted medium to each Petri plate with the diluted sample. Turn each plate gently to mix the contents and spread the cells throughout the medium.
- Let the plates settle down.
- At 37°C, incubate these plates upside down for 24 to 48 hours.

RESULT

Select the plates that have between 30 and 300 colonies produced. Plates with more than 300 colonies are too numerous to count (TNTC) because it is impossible to count them and plates with less than 30 colonies are too few to count (TFTC). Only count plates with between 30 and 300 colonies on them. Don't forget to count both surface and subsurface colonies.

Multiplying the number of colonies counted by the dilution fac tor yields the number of organisms per milliliter of the original culture:

The number of cells per ml = the number of colonies/dilution factor.

OBSERVATION AND RESULT

Physical parameters

Table 1: Temperature of the sample

Sample name	Temperature (in degree celcius)
Sampling 1	32
Sampling 2	30

Table 2 : sechi depth and colour of the sample

Sample name	Distanc e from observe r to water	Total distance at which disk disappear and reappear	Sechi depth	Sechi colour code
sampling 1	1.08	2.69	1.6	13
sampling2	1.86	2.63	0.76	14

Sample name	weight of the clean plate	weight of the container with water sample	Remain ing weight of contain er after drying	Total dissolv ed solvent
sampling 1 1st sample taken	41.9 g	62.5 g	42.6 g	0.7 g
sampling 1 2nd sample taken	45.0 g	65.4 g	45.8 g	0.8 g
sampling 2 1st sample taken	44.1 g	64.4 g	44.8 g	0.7 g
sampling 2 2nd sample taken	43.3 g	63.7 g	44.1 g	0.8 g

Table 3 : Total dissolved solvent in the sample

Chemical parameters

Table 4 : Nitrate in the given sample (spec reading)

Sample name	Nitrate Reading
standard of	
sample 1	0.174
standard of	
sample 2	0.037
Blank	nullified
sampling 1	-0.055
sampling 2	-0.009

Table 5 : Dissolved oxygen in the sample

Sample name	Dissolved Oxygen
sampling 1	37.6 mg/L
sampling 2	16 mg/L

Table 6 : Primary productivity with light and dark method

Sample name	Dissolved oxygen	Gross primar y produc ti- vity	Net primary product i- vity
Blank	2 mg/L	-10	-20
Sample kept in dark	1.2 mg/L		
Sample kept in light	0.4 mg /L		

Biological parameters

Table 7: Estimation of MPN for the detection of coliform bacteriaon water samples in marine drive

Sample	volume	No. of tubes		MPN /100ml
	inocula ted	colour change	Gas produc ed	
sample 1	10	3	3	
	1	3	3	2400/100ml
	0.1	3	3	
sample 2	10	3	3	
	1	3	3	2,400/100ml
	0.1	3	3	

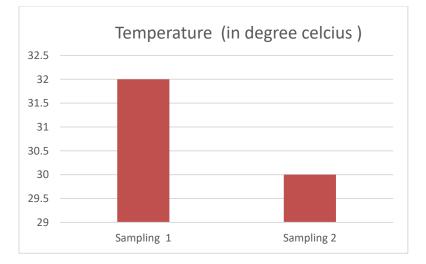


Fig 1: chart for temperature of two sampling

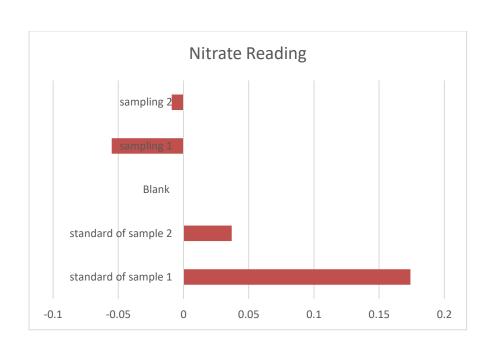


Fig 2: Chart for the nitrate in sample

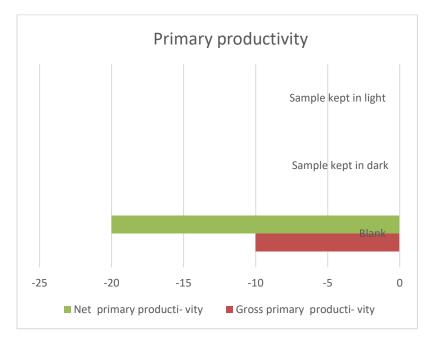


Fig 3: chart for the primary productivity

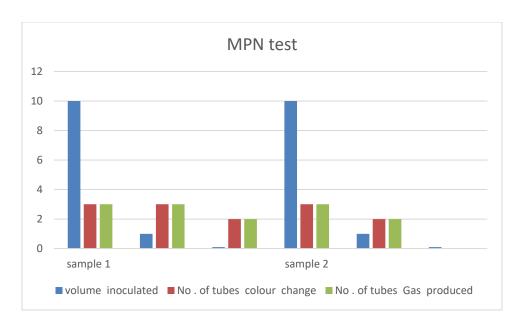


Fig 4: graph for the results of MPN test



Image 2: TDS test – presence of the dissolved solvents in sample collected in petri plate

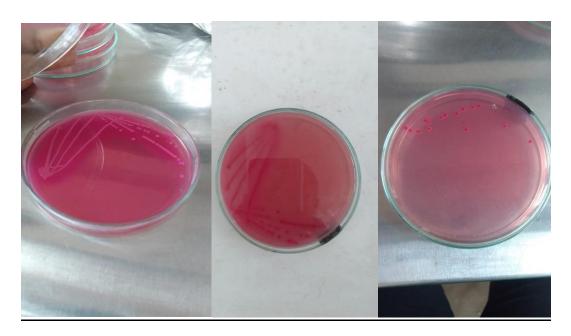
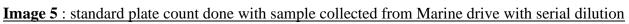


Image 3: completed test of E.coli



Image 4: Nitrate estimation of samples collected from marine drive





DISCUSSION

E –coli in water is a strong indicator of sewage or animal waste contamination. Sewage and animal waste can contain many types of disease causing organisms. Consumption may result in severe illness, children under 5 years of age and those with compromised immune system and the elderly are very suspectible. In India the rate of deaths due to Escherichia coli (E.Coli) per lakh population stood at 16.1 in 2019.

The average temperature from the samples of marine drive is 31°C. (table 1). The average temperature observed during the study of physico-chemical characteristics of vembanad backwaters at Eramalloor region, Alappuzha district, Kerala, India by Ranjith Kumar et.al was 29.0°C. The maximum temperature was observed during the month of May (30.8°C) and the minimum temperature was observed during the month of July (26.2°C). Temperature showed a positive correlation with salinity in that study.

The average dissolved oxygen found in the sampling station of marine drive is 26.8 mg/L. (table 5) The average dissolved oxygen was 4 mg/ml and a low value of 3 mg/ ml was observed in station [1] and highest was observed in station [3]. A DO content of greater than 5 mg/l in water is required for sustaining aquatic Fauna. The dissolved oxygen level in water is constantly changing and represents a balance between respiration and decomposition that deplete oxygen and photosynthetic activity that increases it [8]. Presence of organic waste in water may overload a natural system causing serious depletion of the oxygen supply that in turn leads to fish kills. So we can conclude that the given water from sample station from marine drive area is very suitable for the growth if aquatic organisms.(Yadu Krishnan et.al)

The Most Probable Number is an indicator to presence of coliform bacteria. The presence of coliform bacteria is confirmed by gas bubbles formed in Durgham's tube .Gas bubbles in 9 tubes from the sample collected from marine drive area (Table 7). The presence of fecal coliform bacteria was further confirmed in water samples through completed test of E.coli bacteria on the MacConky agar plates.

The average nitrate was about -0.041 ppm on our study as the value of nitrate in water is very low. The average nitrate was about 0.55 ppm during the study period. Nitrogen, phosphorous and potassium are the results of chemical fertilizers application in the agriculture fields. Nitrate and nitrite content showed a value between a higher concentration of 0.35 and 0.83 and 0.009 and 0.01, for nitrate and nitrite, respectively. Nitrite has got significant correlation (P>0.05) pH content of the water. The permissible limit of nitrate is 50 mg/l. Levels exceeding 0.55 mg/l (ppm) nitrite-nitrogen can cause 'brown-blood' disease in finfish. (Krishnan et.al)

Primary productivity is a rate of changes of biomass per unit area by primary producers at the level of first trophic level. This is done with the help of solar radiation utilizing water and carbon dioxide and sufficient minerals by the process of photosynthesis. In water this productivity is assess by light and dark oxygen bottle method. Results should be like , DO is highest in Light bottle (L) water followed by that of Initial bottle (I) water and lowest in Dark bottle (D) water. But on our results there is lowest DO in the light bottle water and there Iis highest amount of DO in the dark bottle and blank or initial sample bottle water.

Sechi depth of the observed area is almost the same as the water quality and color is observed as not that different from the two times of sample collection .The average Sechi depth of the area is 1.25 m .the color of the water observed is almost at sechi color code of 13 that shows an almost greenish brown color of the water. This identifies the presence of algal blooms , chemicals and other pollutants that are being dumped to the water source. TDS results also show the presence of different contaminants present on the water source used by a lot of people in kerala.

This study has provided a comprehensive analysis of the water quality of Vembanad lake , revealing significant pollution levels attributed . Through rigorous sampling and analysis, we have identified key contaminants exceeding permissible limits set by regulatory bodies, posing potential risks to human health and the environment. Our findings underscore the urgent need for proactive measures to mitigate pollution and safeguard water quality.

CONCLUSION

The purpose of the study was to examine the physicochemical features of Vembanad Lake at Marine drive area in order to assess the lake's overall health and water quality. As part of the procedure, the sample was collected again, one week apart, from the Marine drive area. A procedure that took into account biological, chemical, and physical aspects was used to analyze the material. Among the physical parameters that were measured were the temperature, color, depth of the sechi disk, and total dissolved solvent. Among the chemical measures were nitrate, BOD (biological oxygen demand), and primary productivity. Among the biological criteria are the MPN test, the standard plate count, and the full E. Coli test for the water samples collected at two different periods.

From this results of this study it has been conclude that: The physicochemical analysis of the study revealed that: some of the parameter were beyond the permissible limits .It is estimated from the experiments and observations that water of vembanad lake is under pollution and it is not fit for drinking or domestic purpose .Prior treatment of water is highly important before using it for human purposes .Nitrate testing which was a chemical parameter results that amount of nitrate in surface water of marine drive was significantly less .hence it can it said that nitrate is taken up by plants for it is plant nutrient.The value of nitrate is in its permissiable limit and water quality is not compromised because of its presence. From presumptive test and its confirmatory test performed . E- coli presence was observed and was able to isolate it through experiment.This is strong indicator of sewage and animal waste contamination.Consumption of water containing E .coli can lead to compromised immune system and severe illness.

Vembanad Lake and the wetlands around it are experiencing a severe ecological and environmental disaster as a result of human activity and rising pollution. Every day, 2 million people worldwide drink water tainted with excrement. The biggest threat to the safety of drinking water is microbiological contamination caused by fecal pollution. All three stations' worth of water samples had lactose-fermenting bacteria, primarily E. coli, according to the MPN test results. This suggests that there is poo in the water. Fisheries are declining, water quality is deteriorating, there is extensive sedimentation, etc. The management strategies recommended for biodiversity conservation include pollution control, improving water quality through solid and liquid waste management, and regular monitoring. As a result, it is imperative to take a holistic approach to the conservation and management of wetland systems.

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