ASSESSMENT OF MICROPLASTICS CONTAMINATION OF SURFACE WATER AND SOME FISH SPECIES IN OVIA RIVER, BENIN CITY, NIGERIA.

BY

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UNIVERSITY OF BENIN
BENIN CITY

AUGUST, 2025

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AUGUST, 2025

CERTIFICATION

We certify that this work was carried out by Kevwe Precious ORIENVWEN in the Department of Animal and Environmental Biology, University of Benin, Benin City

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CERTIFICATION OF THESIS

We the undersigned attest and declare that the thesis of Kevwe Precious ORIENVWEN titled Assessment of Microplastics Contamination of Surface Water and Some Fish Species in Ovia River, Benin City, Nigeria, has successfully passed the anti-plagiarism test and does not violate any copyright regulations.

Prof. M. O. Omoigberale **Date**
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Prof (Mrs.) I. Tongo **Date**
(Head of Department)

DEDICATION

This work is dedicated to Yahweh, who is God Almighty.

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ABSTRACT

Microplastics have become an emerging pollutant of concern in aquatic environments due to their persistence, potential toxicity, and ability to bioaccumulate in aquatic organisms. However, limited information exists on the extent of microplastics contamination in Nigerian freshwater systems, especially in the Ovia River. This study aims to assess microplastics contamination in surface water and some fish species (*Notopterus afer* and *Heterobranchus bidorsalis*) of the Ovia River in Benin City, Nigeria, with the goal of determining pollution levels, potential sources, and associated health risks.

Data were generated through the collection of water and fish samples from four stations along the river from January to June, 2024. Physicochemical parameters of the water were measured using standard procedures, and microplastics were extracted from both water and fish digestive tracts using density separation and wet peroxide oxidation methods. Polymer types were identified using Fourier Transform Infrared Spectroscopy (FTIR), while elemental composition was analyzed using Energy Dispersive X-ray Spectroscopy (EDX). Particle size distribution was examined using Scanning Electron Microscopy (SEM), and ImageJ software was used for image analysis. Pollution indices such as Water Quality Index (WQI), Contamination Factor (CF), and Carcinogenic and Non-carcinogenic Risk Indices (CDI, HQ, HI, CR) were also applied.

The analysis of physicochemical parameters in the Ovia River revealed generally acceptable levels with no significant differences ($P > 0.05$) across the study stations, except for copper, chromium, and manganese which showed significant variation ($P < 0.05$). Heavy metal concentrations were ranked: Fe > Zn > Cr > Cu >Mn > Ni > Pb, while arsenic and vanadium were below detection limits. Water Quality Index (WQI) and Comprehensive Pollution Index (CPI) classified Station 1 as having good water quality, while others were rated poor. Health risk assessment using Chronic Daily Intake (CDI) identified iron as the most prominent exposure risk. SEM revealed spatial variation in microplastic particle sizes in both water and fish, with the highest mean sizes observed in station 2 and in *Notopterus afer* respectively. Polymer analysis identified polyvinyl alcohol and nylon as the dominant microplastics in surface water, while LDPE (Low-density polyethylene) and polypropylene were most prevalent in fish samples. However, *Heterobranchus bidorsalis* exhibited a more diverse and complex microplastics polymer profile, indicating greater ingestion and exposure compared to *Notopterus afer*. The elemental composition in water and fish samples identified Cl, N, Na, C, Ca, Mg, Si, P, K, Al, Fe, S, Ti, Ag, and Mn as additives to microplastics with high intensity peaks of chlorine and carbon. Overall, the Ovia River ecosystem is moderately polluted with microplastics, and there is evidence of bioaccumulation in fish, necessitating urgent environmental management strategies.

CHAPTER ONE

INTRODUCTION

1.1 Background to the study

Since the 1950s, the production of plastics has increased rapidly due to the development of large-scale industrial manufacturing. Plastics are used in almost every aspect of daily life, including packaging, building and construction, automotive, electrical and electronic, agricultural, and consumer and home appliances. Plastics are also used in sport, health, and safety applications. Microplastic pollution has emerged as a global environmental issue with widespread distribution in both freshwater and marine ecosystems (Zhang *et al*., 2016). In recent years, there has been increasing attention to the presence and effects of microplastics in freshwater environments, including surface waters and sediments (Eerkes-Medrano *et al*., 2015; Vaughan *et al*. 2017).

Microplastic (MPs) are small plastic particles less than 5mm in size that can travel long-distance via circulation of the water or sea surface due to their buoyant properties (Density 0.917-1.61 gcm-^{3}) and prevalent nature (Thevenon and Carroll, 2015; Campbell *et al*., 2017; Wu *et al.,* 2019), that are causing increasing concerns due to their widespread presence in the environment, including in rivers, lakes, and oceans. These plastic particles come from a variety of sources, including the breakdown of larger plastic items such as plastic bottles and bags, as well as microbeads in personal care products and synthetic clothing fibers. These particles are found extensively in the environment due to their widespread use and persistence. Owing to their small size, microplastics can be ingested by a variety of organisms and have been detected in numerous species of fish and invertebrates, raising concerns regarding their safety for human (Lusher *et al*, 2017)

Interestingly, while microplastics are a contemporary focus, traditional pollutants such as heavy metals have been extensively documented in the Ovia River and its biota, with some levels exceeding recommended limits for drinking water and fish (Omoigberale and Ogbeibu, 2005; Isibor *et al.,* 2016). This suggests a multifaceted pollution problem affecting the river's ecosystem. Moreover, the Ovia River is not only a source of drinking water but also receives urban drainage and agrochemical wastes (Imoobe and Adeyinka, 2010), which could exacerbate the contamination levels.

In general, microplastic are classified into primary microplastics and secondary microplastic. Primary microplastic include any plastic fragments or particles that are already 5.0 mm in size or less. These primary MPs are derived from microbeads in cosmetics, cleaning products, and air-blasting media, which can directly enter the environment (Du and Wang, 2021). The secondary microplastic are derived from the decomposition of larger plastic pieces, this typically happens when larger plastics undergo weathering, through exposure to wave action, wind abrasion, and ultraviolet radiation from sunlight. Such sources of secondary microplastic include water and soda bottles, fishing nets, plastic bags, microwave containers, tea bags and tire wear (Boucher and Friot, 2017; Conkle *et al*. 2018; Kovochich *et al*. 2021). Pollution of microplastic has gained significantly more research attention in recent years due to its persistence in the natural environment and potential adverse impacts on organisms. Microplastics can migrate in various environmental compartments, such as air (Dris *et al*. 2016), soil (Blaesing and Amelung, 2018), oceans (Andrady, 2011), and freshwater (Dris *et al.* 2015). The entry of microplastics into the environment can occur through various pathways, which can differ across different geographical regions. For example, primary MPs used in consumer cosmetics are probably more important in affluent regions (Lambert, 2013). Choudhury *et al*. (2018), stated that microplastics have several potential environmental release pathways which are; passage through waste water treatment plan (WWTPs), either from MP use in personal care products or release of fibres from textiles during the washing of clothes, to surface waters, application of biosolids from WWTPs to agricultural lands, storm water overflow events, incidental release (e.g. during tyre wear), release from industrial products or processes, and atmospheric deposition of fibres.

Microplastic have been found to have detrimental effects on aquatic organisms, including fish, by blocking their digestive systems, releasing toxic chemicals, and disrupting hormone levels (Malik *et al*., 2021). Many organisms swallow pieces of plastic, which can accumulate in their digestive system (Matsuguma *et al*. 2017). As documented by Ryan (2016), crustaceans can accidentally swallow plastic during secondary ingestion, which occurs when they feed on prey that has previously ingested plastic debris. While microplastic affect a wide range of aquatic organisms, fish are particularly vulnerable due to their high levels of microplastic consumption. The deliberate ingestion of microplastic by fish has been observed, as fishes often mistake these tiny plastic particles for suspended food particles in the water or unintentionally when they ingest it along with food (Egbeocha *et al*. 2018). Espinosa *et al*. (2018) found that continued exposure of fish to Polyvinyl Chloride (PVC) or polyethylene (PE) could destroy fish immune system due to the oxidative stress in the fish leucocytes. Specifically, PVC showed more changes in fish leucocytes than PE. Once organisms ingest MPs, they may experience serious physical and toxicological effects. These effects are categorized as physical (related to the shape, color, and size of the particles) and chemical (related to the presence of additives and/or absorbed chemical contaminants. For instance, some parameters such as the level of contamination, location and local human activity are important in assessing MP accumulation in fish muscles (Akhbarizadeh *et al*. 2018). The ingestion of microplastic leads to several physical effects, including the blockage of alimentary appendages and the digestive system, ultimately disrupting the balance of the entire ecosystem. Additionally, microplastic can also pose a threat to human health as they can accumulate in the tissues of fish and other marine organisms that are consumed by humans (Alberghini *et al*. 2022).

Ovia River is a major water body in Benin City, Edo State, Nigeria, and is a significant source of water for the local community as well as supporting a variety of flora and fauna. The Ovia River is located in southern Nigeria and ranks among the most polluted river systems in Nigeria (Edoreh *et al*. 2020). Tongo *et al. (*2017) states that, the river has played a crucial role in the sustenance of the densely populated southern Nigerian community, but in recent years, it has been under great risk due to unregulated human activities such as illegal mining, land runoff, and waste dumping. Tongo and Ezemonye (2018), carried out an environmental assessment in 2005 and revealed sharp increase in the concentration of heavy metals and hydrocarbons in the river and in the sediments due to human activities. The contamination of the river by metals and hydrocarbons has been assessed in various parts of the Ovia River to have come from all the anthropological sources listed above. Ibrahim *et al*. (2023) observed that in Nigeria today, there is an increase in the use of plastic-based packaging such as bottles, containers, and carrier bags. The increase in production of these plastic materials has increased the probability of plastic particles getting into water bodies. However, there is limited information on the levels of microplastics contamination in inland river and its potential impacts on water quality and fish populations (Doherty *et al*. 2024).

The Ovia River, like many freshwater systems globally, faces the threat of microplastics contamination, with potential implications for aquatic biota such as *Heterobranchus bidorsalis*. While research on microplastics in Nigerian rivers is evolving, there is a critical need for comprehensive studies focusing on the Ovia River to assess the extent of contamination, identify sources, and evaluate its impact on aquatic organisms. Such investigations are essential for informing effective management strategies to mitigate microplastic pollution and safeguard the ecological integrity of freshwater ecosystems in Nigeria (Egbuna *et al*. 2025).

1.2 Statement of Research Problem

Microplastic contamination in aquatic environments has become a major global concern due to its persistence, potential for bioaccumulation, and capacity to carry harmful pollutants. While marine ecosystems have received significant attention, freshwater systems especially in Nigeria remain underexplored.

Recent studies have confirmed the presence of microplastic in Nigerian rivers and estuaries, including water, sediments, and fish (Attah *et al*. 2023). However, comprehensive data on their distribution and impact in key freshwater systems like the Ovia River are lacking. This river, which supports domestic use and diverse aquatic life such as *Heterobranchus bidorsalis* and *Notopterus afer* may be at risk from unmonitored microplastic pollution.

Freshwater bodies are increasingly recognized as pathways for microplastics to marine environments (Sighicelli *et al*. 2018), yet research focusing on local rivers and associated biota is limited. Without baseline data on the abundance, types, and distribution of microplastic in the Ovia River, effective management and mitigation strategies cannot be properly developed.

1.3 Justification of Study

The assessment of microplastics contamination in surface water and fish in the Ovia River, Benin City, Edo State, is a crucial study that addresses a growing environmental concern with significant implications for both ecosystems and human health. Microplastics, defined as plastic particles smaller than 5 mm, have become ubiquitous in aquatic environments, posing serious risks to aquatic organisms and the food chain (Smith *et al*., 2018). Investigating their prevalence in the Ovia River is especially pertinent given the river's role as a vital resource for the surrounding communities, providing water for domestic use, irrigation, and fishery.

Understanding the extent of microplastic contamination in this specific river will not only contribute to the body of knowledge regarding environmental pollution in Nigeria but also serve to highlight the unique challenges faced by developing regions. The socio-economic factors that contribute to plastic pollution, such as rapid urbanization and inadequate waste management practices are pertinent to the study site (Auta *et al*., 2017). Therefore, this research serves not merely as an academic exercise but as a means to advocate for improved local environmental policies and practices.

Furthermore, the ramifications of microplastics ingestion by fish species in the Ovia River can lead to bioaccumulation of toxic substances and have the potential to impact human health through consumption. With fish being a staple protein source in the diet of local populations, it is crucial to assess the risks associated with microplastics contamination in aquatic life and its implications for public health (Rochman *et al*., 2013). The findings from this study could prompt public awareness initiatives and provide empirical evidence to support the implementation of better waste management strategies.

Additionally, this research will generate baseline data that could pave the way for future studies, enabling long-term monitoring of microplastics levels in both water and fish populations in the region. Given the limited research on microplastics in freshwater systems within Africa, this thesis will contribute significantly to filling this knowledge gap, aiding in the development of a comprehensive understanding of microplastics pollution and its environmental impacts (Jambeck *et al*., 2015). The study's outcomes have the potential to influence policy-making and foster community engagement aimed at mitigating plastic pollution, thus promoting sustainable practices in the Ovia River basin.

1.3 Aim and Objectives

The aim of this study was to access the levels of microplastic contamination in surface water and some fish species in Ovia River, Benin City, Nigeria.

The objectives of the study were to:

i. assess the spatial and temporal variations in the concentrations of the physical and chemical parameters including the heavy metals of the surface water of Ovia River.
ii. determine the type, abundance, distribution and the spatio-temporal variations of microplastics in the surface water of Ovia River.
iii. examine the presence, size distribution and types of MP polymers in some fish species (*Notopterus afer* and *Heterobranchus bidorsalis*) in the Ovia River, Edo State, Nigeria.

CHAPTER TWO

LITERATURE REVIEW

Aliyu *et al*. (2023) analyzed MP concentrations in the Kaduna River, treated water from two conventional water treatment plants, brands of bottled water, and food-grade salts available in Kaduna Metropolis, Nigeria using standard methods. Data obtained show that levels of MPs ranged from 25 to 36 particles L^{-1} in treated water, and to 153 particles L^{-1} in the river. Samples of bottled water contained 1.4 to 3.7 particles L^{-1} and samples of table salt also contained 0.13 to 0.27 particles g^{-1}. Water and salt samples contained five different types of polymers, including polyethylene, polypropylene, polyester, polyvinyl chloride and polyethylene terephthalate. Additionally, MPs were divided into three groups based on their physical characteristics. In both raw and treated water, fragments were clearly more prevalent; bottled water and in table salt, fragments and fibers predominated. Microplastics in the bottled water posed a medium pollution risk, according to pollution risk indices. The estimated daily intake was generally minimal, indicating little harm from daily consumption, but it also demonstrates that children have a larger intake of microplastics than adults. Leaching from the packing material was identified as the MPs' primary source.

The presence of plastic debris in oceans poses a significant concern due to the potential accumulation of harmful chemicals, which raises important questions about the extent to which microplastics in seafood contribute to human exposure to these compounds. These chemicals can originate from the plastic itself or be absorbed from the surrounding environment. Plastics, with their hydrophobic surfaces, have a propensity to adsorb compounds with similar chemical properties from seawater. Persistent organic pollutants (POPs) like polychlorinated biphenyls

(PCBs), dichlorodiphenyltrichloroethane (DDT) and its metabolites dichlorodiphenyldichloroethane (DDD) and dichlorodiphenyldichloroethylene (DDE), hexachlorocyclohexanes (HCHs), and polycyclic aromatic hydrocarbons (PAHs) are among the substances of concern. Studies, such as that conducted by Mato *et al*. (2001) which examined the concentrations of PCBs and DDE on plastic resin pellets collected off the coast of Japan have shown levels up to 105–106 times higher than in the surrounding seawater. Similar findings have been reported in plastics sampled from oceans and beaches worldwide. These lipid-soluble chemicals are well-recognized concerns in human nutrition, prompting close monitoring of their levels in food, often with established threshold values (Clemence and Gurerrant, 2016).

Tibbetts *et al.* (2018) investigated microplastic contamination in a heavily urbanized river system, focusing on the River Tame and four of its tributaries in Birmingham, United Kingdom. Sediment analysis revealed that all sites contained microplastic, with an average concentration of 165 particles per kilogram, making the catchment one of the most contaminated urban rivers reported at the time. The study showed that although urban environments generally had higher levels of microplastic compared with rural settings, the abundance of particles did not follow a straightforward relationship with population density or proximity to wastewater treatment facilities. Instead, hydrological features played a stronger role: the presence of a lake along the course of the River Tame significantly reduced flow velocity, encouraging deposition of fine sediments and microplastics. This highlighted the importance of in-channel and floodplain dynamics in shaping microplastic distribution. Their findings underscore that urban rivers serve as critical sinks and transport pathways for microplastic, influenced not only by anthropogenic inputs but also by hydrodynamic and ecological processes.

Campanale *et al*. (2020) investigated the abundance and composition of microplastic in the most important river of Apulia Region (Southeast Italy) evaluating the main drivers and possible input sources of microplastic debris. Microplastic concentrations ranged from 0.9 ± 0.4 particles/m^3 to 13 ± 5 particles/m^3 showing comparable values to or greater than those ones reported in other studies (Yuan *et al*. 2021; Gabriel *et al*. 2023). A significant difference ($p < 0.05$) in the average microplastic concentrations in different campaigns of the study was observed, suggesting a temporal variation in plastic abundances. These could be explained by the hydrology of the river that influences the particle concentration with its physical properties such as flow velocity, water level and seasonal variability. Microplastic were found at higher concentrations during wet periods indicating a land-based origin probably connected to waste produced by the surrounding agricultural areas. In fact, there was a strong statistically significant positive correlation between the concentration of microplastic and the water level ($R = 0.8475$, $p < 0.0001$) was observed.

Cheung *et al*. (2019) aimed to quantify microplastic abundance and the properties of the microplastic at the surface of an urban river after a rainfall event in Hong Kong. Plastic samples were collected at the river surface after a 3-day rainfall event. The microplastic abundance was 7.428 pieces/m^3 (1,615,506 pieces/km^2), which was nearly double of that observed on coastal sea surface of the same area (3.973 pieces/m^3). Microplastic abundances were found to decrease over ten times (from 14.015 to 1.298 pieces/m^3) within 2 hours, suggesting that the temporal distribution of river microplastics was highly dynamic after rainfall.

Mani *et al*. (2018) investigated the microplastic profile along the Rhine River, one of the largest European rivers. Measurements were made at 11 locations over a stretch of 820km. Microplastic were found in all the samples, with 892,777 particles km^{-2} on average. In the Rhine-Ruhr metropolitan area, a peak concentra6tion of 3.9 million particles km^{-2} was measured.

Microplastic concentrations were diverse along and across the river, reflecting various sources and sinks such as waste water treatment plants, tributaries and weirs. Long-term monitoring studies have revealed temporal trends in microplastic pollution, including seasonal variations and trends over time. For example, fluctuations in microplastic abundance have been linked to changes in hydrological conditions, such as rainfall patterns and river flow rates (Sadri and Thompson, 2014; Imhof *et al.* 2016). Understanding these temporal dynamics is essential for assessing the effectiveness of management measures and predicting future trends in microplastics pollution.

Wang *et al.* (2019) studied the degree of microplastic pollution in surface waters of Lake Ulansuhai, a major freshwater lake in the Yellow River basin of northern China. Results showed microplastic concentrations ranging from 1760 ± 710 to 10,120 ± 4090 particles per cubic meter of water (n/m^3). The microplastic spatial distribution is heterogeneous, with higher levels near the drainage canal entrance of Lake Ulansuhai, and a downward trend from north to south in the lake. The main type of microplastics is colored particles, including fibers as the most abundant. More than 80% of microplastics were less than 2 mm. Fourier Transform Infrared Spectroscopy (FTIR) analysis results showed that the main plastics were polyethylene, polystyrene and polybutylene terephthalate. There were also some metallic elements adsorbed on the surface of microplastic, such as Fe, Ca and Zn, detected by energy-dispersive spectrometry. The presence of metallic elements may worsen water pollution.

Wang *et al.* (2020) assessed occurrence and pollution characteristics of microplastic in surface water of the Manas River Basin, China. The range of abundance of microplastic in the Manas River Basin was 21 ± 3 – 49 ± 3 items/L. Fibrous microplastic were dominant in all the sites (88.0%); their size was mainly distributed between 0.1 and 1.0 mm (82.6%), and white and black were the dominant colours (82.9%). In addition, the size range of flaky-type microplastic was investigated in the study, which was principally between 2.5×10^3–9.0×10^4 μm^2 (84.5%). Infrared spectral analysis revealed that most of the selected particles were identified as microplastics, and polymer types of microplastic were dominated by polypropylene and polyethylene terephthalate (48.3%).

Saipolbahri *et al*. (2020) conducted an investigation to determine the presence of microplastic in surface water of Kelantan Bay, Malaysia. The samples were collected between May and July 2019. Microplastic were isolated from the surface water and sediment samples by using wet peroxide oxidation (WPO) followed by density separation. Microplastic were sorted visually according to their shapes and colours after being examined under photographed microscope. The most abundant particle found in this study was threadlike and fragment shapes. Fourier transform infrared (FTIR) spectroscopy was used to identify functional groups in the composition of microplastics to determine the types of microplastic found in the samples. Polypropylene (PP) was the most abundance type of microplastic found in the surface water and sediments of Kelantan Bay, Malaysia. This study provides an insight of the types of microplastics found in Kelantan Bay and therefore, further actions need to be taken to curb the distribution of microplastic in marine ecosystem from threatening the marine food chain of Kelantan Bay.

He *et al*. (2020) investigated microplastic contamination of the Yangtze River from the upper reaches to the estuary using different sampling methods to understand extensive information on microplastic pollution. The results showed that the average abundance of microplastic ranged from $1.62 \pm 0.61 \times 10^5$ to $4.25 \pm 3.87 \times 10^6$ items/km^2 (trawling samples) and 800.0 ± 300.0 to 3088.9 ± 330.6 items/m^3 (filtering water samples). The average abundance (by trawling) in the Three Gorges Reservoir ($2.80 \pm 1.86 \times 10^6$ items/km^2) was one order of magnitude higher than that of other sections, which affirmed the barrier effect of dams on microplastic distribution. The barrier effect was more obvious on larger size particles when the results of two methods were compared. The abundances near the left, right banks and in the midstream showed no significant difference when both methods used were compared. Characteristics analysis demonstrated that fragments (47.9%) dominated in trawling samples and fiber (63.4%) dominated in filtered water samples. Microplastic of small sizes (<1 mm) were transparent and dominant in samples collected by the two methods. Polyethylene (PP) and polypropylene (PE) were the dominant polymer types in the detected microplastic.

Li *et al*. (2019) investigated the correlation between lake trophic state and microplastic abundance in surface water and sediments in Yangtze River (China). The microplastic abundance in surface water was approximately two orders of magnitude lower than that in sediments in all 18 lakes. Hong Lake had the highest microplastic abundance in surface water sample and the dominant microplastic shape was fiber of 93.81% in surface water sample. Blue-colored microplastic were dominant in nearly all lakes in surface water samples (around 40%–60%) and sediment samples (around 60%–80%), followed by purple- and green-colored ones. The microplastic size <1 mm was dominant in surface water sample (around 40%–60%) and sediment sample (around 50%–80%). The dominant polymer was polypropylene in surface water samples (around 60%–80%).

Murphy *et al*. (2022) assessed the level of microplastic in Irish river sediment. A total of 690 MPs were recovered from the 16 sites, with fibres as the dominant MP type. The highest concentration of MP was 155 MP fibres/kg wet sediment found in samples, collected from Graiguenamanagh, Co. Kilkenny (GK). The majority of the recovered MPs were polyethylene (PE), polypropylene (PP), nylon, and cellulose acetate (CA) fibres. Overall, their study highlighted the presence of MP in Irish river sediments and provided a baseline for future studies on MP pollution.

Nel *et al*. (2018) assessed microplastic dynamics in the Bloukrans River catchment in the Eastern Cape, South Africa, focusing on seasonal variations in an urban river system. Sediment and *Chironomus* spp. larvae were analyzed across summer and winter, revealing higher microplastic concentrations during the winter when reduced flow promoted sediment deposition. Distribution patterns were also linked to substrate type and organic matter content. Importantly, the study showed that benthic deposit feeders ingested microplastic, with *Chironomus* larvae reflecting seasonal changes in sediment contamination. The findings suggest that such organisms can serve as biological indicators of microplastic pollution in degraded freshwater environments.

Several studies have investigated the relationship between physicochemical parameters and microplastic contamination levels in water bodies. Factors such as pH, temperature, dissolved oxygen, and turbidity have been found to influence the distribution and abundance of microplastic in aquatic ecosystems (Eerkes-Medrano *et al*. 2015; Zhang *et al*. 2020). Understanding these relationships can provide insights into the transport and fate of microplastics in the environment.

Park *et al*. (2020) investigated the distribution of microplastic in surface water, fish, and sediment near a sewage treatment plant (STP) in the Tanchon stream, one of the main tributaries flowing into the Han River, Korea. The concentration of microplastic in water varied spatially and temporarily, ranging between 5.3 and 87.3 particles/m^3 (31.4 ± 28.5 particles/m^3). In fish, the concentration in upstream and downstream sites was 7.3 ± 7.3 and 12.4 ± 17.9 particles/fish, respectively. Spatially, the downstream site was the most polluted with microplastic in water and fish. The concentration of microplastic was positively correlated with fish body length and weight. The contribution of upstream to the microplastic load in downstream was 15.8% in dry season (April), which was higher than that of STP effluent and Yangjaechon creek. Meanwhile, the highest load was observed in STP effluent (5.1%) in rainy season (August). Microplastic were more abundant in the water in the rainy season (37.4 ± 37.0 particles/m^3) than in the dry season (28.2 ± 22.2 particles/m^3). Polyethylene (49%) and polypropylene (18%) were the most abundant polymer types in water and fish. Regarding shape of microplastic, fragments were dominant (95%) over fiber and film in water, fish, and sediment.

Numerous studies have documented the ingestion of microplastic by fish species inhabiting contaminated aquatic environments (Rochman *et al*. 2015; Koelmans *et al*. 2016). Fish can ingest microplastic directly through feeding or indirectly through the consumption of contaminated prey items. Once ingested, microplastic can accumulate in the gastrointestinal tract and translocate to other tissues, potentially causing physiological and behavioral disturbances in fish.

Liu *et al*. (2021) assessed distribution of microplastic in water and fish of the Dafeng River, a remote river in China. During the dry and rainy seasons, the microplastic content of the surface water ranged from 3×10^{-4} -2.5×10^{-3} items/L (7×10^{-4} -0.12 items/m) and 4×10^{-5} -9×10^{-4} items/L (2×10^{-3} -2.8×10^{-2} items/m^2). The pollution level during the dry season was approximately two to three times higher than that during the rainy season ($P < 0.05$). The estimated annual load of microplastic carried by the Dafeng River flow was 8.3 × 108 particles. The microplastic pollution in the Dafeng River was closely related with residential activities. The contents of microplastic in the digestive tracts and gills of fish ranged from 8×10^2 to 5.7×10^3 items/kg (0.3–6.7 items/individual) and 2×10^2 to 1.7×10^3 items/kg (0.1–3.0 items/individual), respectively. The proportion of blue particles in fish was significantly higher than that in the water and sediment, which may indicate that they prefer blue-colored items. The microplastic pollution levels in the water, sediment, and fish of the Dafeng River decreased in the following order: fish > surface water in terms of items/kg.

Bakir *et al*. (2020) documented the levels of microplastic in three commercially important small pelagic fish species in South African waters, namely European anchovy (*Engraulis encrasicolus*), West Coast round herring (*Etrumeus whiteheadi*) and South African sardine *(Sardinops sagax*). Data suggested variation between species with a higher concentration of microplastic for *S. sagax* (mean of 1.58 items $individual^{-1}$) compared to *E. whiteheadi* (1.38 items$individual^{-1}$) and *E. encrasicolus* (1.13 items $individual^{-1}$). The occurrence of microplastic was also higher for *S. sagax* (72%) and *E. whiteheadi* (72%) compared to *E. encrasicolus* (57%). Microfibers accounted for 80% of ingested microplastic (the remainder were plastic fragments) with the main ingested polymers being poly(ethylene:propylene:diene) (33% occurrence), polyethylene (20%), polyamide (20%), polyester (20%), and polypropylene (7%). The abundance of ingested items was not significantly correlated with fish caudal length or body weight, and spatial investigation indicated an increase in the abundance of ingested items from the West to the South coast. *Etrumeus whiteheadi* is proposed as a bio-indicator for microplastic for South Africa.

Loayza *et al*. (2018) evaluated the presence of microplastic in the stomach content of four fish species (*Orestias luteus, Orestias agassizii, Trichomycterus dispar*, and *Odonthestes bonariensis*; N = 1,283) targeted by local fisheries in Lago Menor of Lake Titicaca, Bolivia, and looked for relationships with trophic guilds or fishing areas. Additionally, surface water was analyzed to evaluate the presence of microplastic in the water. The results revealed that the frequency of microplastic ingestion was low in all species. Research on the spatial distribution of microplastic in rivers, lakes, and oceans has highlighted the variability in contamination levels across different sampling sites. Factors such as proximity to urban areas, industrial activities, and wastewater discharge points have been identified as significant contributors to microplastic pollution in freshwater systems (Lebreton *et al*. 2017; Horton *et al*. 2018). Mapping the spatial distribution of microplastic can help identify hotspots of contamination and prioritize conservation efforts.

Ghosh *et al*. (2020) investigated the abundance and characteristics of microplastic in commercial fish species from the Bay of Bengal, off the coast of Bangladesh. The gastrointestinal tracts of 100 individuals from 10 species of commercial marine fish were examined for microplastic. Microplastic were detected in all species. A total of 215 microplastic were recovered with an average abundance in the range of 2.2 ± 0.89 microplastic per individual. Five types of microplastic were found: fibers, films, fragments, foams, and granules, which accounted for 53.4%, 40%, 3.3%, 1.9%, and 1.4% of the total, respectively. Among the identified microplastics, green (39%) was the most dominant color, and < 500µm (85%) size was the most dominant. Polymers were polyethylene (55%), polypropylene (33%), polyester(6%), polyurethane (2%),ethylene propylene diene monomer (2%), and styrene butadiene rubber (2%).

Rodrigues *et al*. (2019) investigated the contamination of an urban impacted estuary (Douro estuary, NW Portugal) by MPs, including the abundance and distribution of MPs and fish larvae in the estuary. Monthly sampling surveys were performed from December 2016 to December 2017, in nine stations along the estuary. Different types of MPs were found, namely fibers, soft/hard plastic, colorful/transparent plastic, in a total of 2152 particles, with a mean density of 17.06 MPs 100 m^{-3}. Hard MPs and fibers were the most predominant types, representing 83% of the total MPs collected. In some months the number of MPs surpassed the number of fish larvae, with an average ratio of 1.0 fish larvae:1.5 MPs. Such results are concerning, highlighting that a higher availability of MPs may facilitate their ingestion by fish and therefore increase possible impacts in these communities.

Garcia *et al.* (2020) studied the prevalence of microplastic (MPs) in freshwater fish from farmed and natural sources. *Oreochromis niloticus* from aquaculture farms in the Huila region in Colombia, and two local species (*Prochilodus magdalenae* and *Pimelodus grosskopfii*), naturally present in surface waters were sampled. Of the particles identified, fragments were the predominant type in the three tissue types (stomach, gill, and flesh) derived from farmed and natural fishes. MicroFT-IR spectroscopy was conducted on 208 randomly selected samples, with 22% of particles identified as MPs based on spectra with a match rate ≥ 70%. A total of 53% of identified particles corresponded to cellophane/cellulose, the most abundant particle found in all fish. Not all fish contained MPs: 44% of *Oreochromis* farmed fish contained MPs, while 75% of natural source fish harboured MPs in their tissues. Overall, polyethylene terephthalate (PET), polyester (PES), and polyethylene (PE) were the prevalent MPs found in the freshwater fish. A broader variety of polymer types was observed in farmed fish. The edible flesh part of fish presented the lower prevalence of MPs compared to gill and stomach (gut), with gut displaying a higher frequency and diversity of MPs.

Bessa *et al*. (2018) assessed the occurrence of microplastic ingestion in three commercial fish species: the sea bass (*Dicentrarchus labrax*), the seabream (*Diplodus vulgaris*) and the flounder (*Platichthys flesus*) from the Mondego estuary (Portugal). Microplastics were extracted from the gastrointestinal tract of 120 individuals by visual inspection and digestion solution. A total of 157 particles were extracted from 38% of total fish sample (96% fibers), with

1.67 ± 0.27 (SD) microplastics per fish. Significantly higher amount of ingested microplastics was recorded for *D. vulgaris* (73%). The dominant polymers identified by μ-FTIR were polyester, polypropylene and rayon (semi-synthetic fiber).

The accumulation of microplastic in fish tissues can lead to a range of adverse effects, including inflammation, oxidative stress, and reproductive dysfunction (Galloway *et al*. 2017; Lu *et al*. 2016). Furthermore, microplastics may act as vectors for the adsorption and transport of chemical pollutants, exacerbating their toxicological effects on fish and other aquatic organisms. Understanding the mechanisms underlying the interactions between microplastics and fish health is critical for assessing the ecological risks associated with microplastics contamination in aquatic ecosystems.

Campbell *et al*. (2020) carried out a study to assess microplastic in the gastrointestinal tracts of fish and the water from an urban prairie creek. Their results revealed microplastics of varying shapes and colours were found in water at all sites in both 2015 and 2016. Concentrations of microplastics were significantly higher at upstream sites (3.8 ± 1.2 and 7.7 ± 2.0 microplastics·m^{-3} in 2015 and 2016, respectively) compared with those at the WWTP OF (1.0 ± 0.2 and 1.0 ± 0.4 microplastics·m^{-3} in 2015 and 2016,) and farther downstream (3.7 ± 0.3 and 0.9 ± 0.3 microplastics·m^{-3} in 2015 and 2016. Results from fish sampled showed that of 181 fish sampled, 73.5% had between 1 and 20 microplastics in their gastrointestinal (GI) tracts, ranging from 50.0% (fathead minnows) to 83.3%. There was a significant difference in the number of microplastics found among species, with northern pike having significantly more compare with other species of fish ($H = 16.487$, $p = 0.002$). Three types of microplastics were observed in both water and fish in Wascana Creek. Although all five species of fish were found to have microplastics in their GI tracts, northern pike had significantly higher values than emerald shiners and fathead minnows.

The gastrointestinal tract from a large number of fish species has been investigated for the presence of microplastic (Barboza *et al*. 2018). Rochman *et al*. (2015) obtained 76 whole fish (11 species) from a fish market in Indonesia and found that 28% of the fish contained plastic debris in the GI tract. They also studied 64 individual fish (12 species) purchased in California and found microplastics in 25%. In a study of 263 fish (26 commercial species) caught off the coast of Portugal, 20% had microplastics in the GI system, and of these, 67% contained one plastic particle (Neves *et al*. 2015). The presence of plastic debris has also been investigated in 290 GI tracts from 3 demersal species (Atlantic cod; *Gadus morhua*, common dab; *Limanda limanda*, and European flounder; *Platichthys flesus*) and 2 pelagic species (Atlantic herring; *Clupea harengus* and Atlantic mackerel; *Scomber scombrus*) in the North Sea and the Baltic Sea (Rummel *et al*. 2016). Plastics, of which 74% were categorized as microplastics, were detected in 5.5% of the samples. Of the 51 mackerel studied, 9 were found to contain plastic debris, while only 1 out of 81 cod had plastic in the GI tract (Gamarro *et al.* 2020).

Foekema *et al*. (2013) reported that none of 84 Atlantic mackerel and only 2% of 566 Atlantic herring caught in the North Sea contained microplastics in the gut. The authors also investigated other species from the North Sea and reported that the number of individuals with ingested plastics was low, and usually only one particle was found. Hermsen *et al*. (2017) examined microplastics in fish from the North Sea taking much care to avoid contamination from external sources and found only 1 out of the 400 GI tracts.

Wesch *et al*. (2017) and Torre *et al*. (2016) have demonstrated that the number of microplastic particles in biological samples easily can be overestimated unless strict methodological precautions are taken. Other studies have found that from 2% to 36% of the investigated fish contained plastics in the guts, and when present, an average number of 2 items or less per fish was detected (Anastasopoulou *et al*. 2013; Bellas *et al*. 2016; Bråte *et al*. 2016; Lusher *et al*. 2013). It has been shown that small indigestible solids or larger microplastics do not accumulate to any large extent in the digestive system and are egested in the feces (Dos Santos and Jobling 1991; Grigoriakis *et al*. 2017; Jovanovic *et al*. 2018). This may contribute to the findings that microplastic are absent in many GI-tracts, and when present, the average number is low. Results from examination of gastrointestinal systems may, however, be of limited relevance to seafood safety, since these organs are not normally consumed (EFSA 2016). Relatively few studies have been carried out on the microplastic content in species that often are consumed whole, e.g. sardines, anchovies, and sprats. Studying European pilchard (*Sardina pilchardus*) and European anchovy (*Engraulis encrasicolus*) caught along the Spanish Mediterranean coast, Compa *et al*. (2018) found that 14–15% had microplastic or natural fibers in the GI tract. Beer *et al*. (2018) investigated the content of microplastic in the digestive system of Atlantic herring and European sprat (Sprattus sprattus) caught in the Baltic Sea during the last 30 years. Independent of the year of harvest, approximately 20% of the fish contained microplastics, and 1 to 2 pieces were detected in the GI tract of these individuals. In another study, only 1–2% of Atlantic herrings and sprats from the Baltic Sea were reported to contain microplastics in the guts (Budimir *et al*. 2018). Sprats and sardines are commonly sold as canned products, and Karami *et al*. (2018) recently investigated 20 different brands of such products for the presence of micro- and mesoplastic (size range 0.149–10 mm). They found that plastics were absent from the fish product in 16 canned brands, while the fish in 4 brands contained 1 to 3 particles per can. It is generally accepted that microplastics of size 0.150 mm (150 µm) or above are not absorbed by the intestine (Lusher *et al*. 2017), and it was, therefore, suggested that plastic pieces in the cans must come from GI tracts present or from contamination during the canning process. Small pelagic species are usually beheaded, and sardines are often gutted prior to canning, while the smaller sprats are commonly kept alive in net pens for at least 48 h after harvesting to empty the intestinal tract before processing (Warne, 1988).

Nan *et al*. (2019) worked on the microplastic occurrence and types in water samples and the shrimp *Paratya australiensis*, and studied if the shrimp could be a suitable bioindicator for microplastic pollution. Surface water and *Paratya australiensis* across ten urban and rural freshwater sites in Lake Victoria were sampled. Microplastic were present in the surface waters of all sites, with an average abundance of 0.40 ± 0.27 items/L. A total of 36% of the shrimp contained microplastic with an average of 0.52 ± 0.55 items/L (24 ± 31 items/g). Fibre was the most common type, and blue was the most frequent colour in both water and shrimp samples. The dominant plastic types were polyester in water samples, and rayon in shrimp samples. Though results from the study showed a relatively low concentration of microplastics in water samples in comparison with other studies (Kroon *et al*, 2018: Leterme *et al*, 2023), they emphaized that microplastics were regularly detected in fresh waterbodies in Lake Victoria, Australia.

Zin *et al*. (2021) studied on the prevalence of MPs in green mussels and their environment is now being questioned as to whether aquaculture practices or human environmental activities cause them. Hence, this study focuses on looking into the prevalence of microplastic (MP) in natural environments and mariculture mussels (*Perna viridis*) which comprise physical characteristics to compare microplastics (levels, type, size and colour) uptake based on the mood of living from Pasir Putih estuary in Johor, Malaysia. Specimens of *Perna viridis* were obtained from two sampling stations in the Pasir Putih estuary. In cultured *P.viridis* samples, microplastic abundance ranged from 0 to 6.67 Mp particles/ individual with average values of 2.23 ± 1.04 MPs particles/individual and 0.4 ± 0.24 for each gram of soft tissue. Meanwhile, the microplastic found in wild *P. veridis* averages 1.29 ± 1.19 Mp particles/individual and 0.44± 0.34 Mp particles/gram. The shapes of microplastics found in the tissue of the green mussel are high in fragments. Most polymers in fragments found in the natural and wild mussels were standard plastic used in plastic products such as polyethylene (PE) and polypropylene (PP).

Primus and Azman (2022), studied the quantification and characterization of microplastic in fish and surface water at Melayu River, Johor. The amount of ingested microplastic by Melayu River fish samples was in the order: Gray Eel-Catfish (3.92 ± 4.17 particles/individual) > Sagor Catfish (2.00 ± 1.41 particles/individual) > Spotted Sicklefish (2.00 ± 0.00 particles/individual). The trend of microplastic by month in water samples was Mar-20 (2.89 ± 1.36 particles/L) > Feb-20 (1.33 ± 1.00 particle/L) and Jan-20 (1.00 ± 0.87 particle/L). Microplastic were mostly in the class size 0 µm – 0.50 µm. In the fish samples, fibres were found to be dominant, while in the water samples, films were dominant. Ingestion of microplastics by colour was ranked as blue > black > red > yellow in fish samples whereas microplastics' colour in water samples was ranked as blue > red > black > translucent > green. Therefore, it was concluded that the abundance of blue microplastics in fish samples was due to the common blue plastics used by the locals. The identified microplastics were of polyethylene terephthalate (PET) and polyethene (PE) origins.

Pan *et al*. (2021) studied the occurrence of microplastic in the gastrointestinal tract and gills of fish from Guangdong, South China. The results revealed that Most of the microplastics were less than 1 mm, white and blue fibers. The abundance, type, shape and color of microplastics in gills and in the gastrointestinal tract (GIT) of eight species of commercial fish in the coastal waters of central and western Guangdong Province were analyzed. The overall abundance of microplastic particles in fish was 6.6 items/individual, with an average of 2.2 particles in gills and 4.4 in the GIT. The GIT of carnivorous fish was less likely to ingest microplastic than that of herbivores or omnivores. Middle-water fish, on the other hand, are less likely to have their gills contaminated with microplastics.

Patidar *et al*. (2024) conducted an investigation of microplastic contamination in the gastrointestinal tracts of different freshwater fish species from the Chhota Nagpur zone in eastern India. A total of 144 microplastic were found in the collected samples of fish species. Among the analyzed samples of MPs, 69.4% were fibers, films were 22.3% and 8.3% were fragments. Polyamide (nylon), polypropylene, polyvinyl chloride, polycarbonate, polyethylene terephthalate and polyethylene polymers were found.

Heterobranchus bidorsalis, commonly known as catfish, is a commercially important fish species in Nigeria's freshwater ecosystems, including the Ovia River. As a benthic feeder, it is susceptible to ingesting microplastic present in sediments and the water column, potentially leading to adverse physiological effects, bioaccumulation, and transfer of contaminants along the food chain (Rochman *et al*., 2013). While limited studies have specifically examined the interaction between microplastic and *Heterobranchus bidorsalis*, analogous research on other freshwater fish species suggests potential implications for their health, reproductive success, and population dynamics (Murphy *et al*., 2016).

CHAPTER THREE

MATERIALS AND METHODS

3.1 DESCRIPTION OF STUDY AREA

3.1.1 Geographical Location

The geographical location of the study area is the Ovia River in Benin City, Edo State, Southern Nigeria. Originating from the Akpata hills in Ekiti State, this significant watercourse traverses through Ovia South-West Local Government Area. Situated within the low-lying terrains of the Niger Delta, the river spans from Latitude 06°23'42.76"- 06°27'10.15"N to Longitude 05°25'55.0"- 05°29'36.20"E. Serving as a crucial tributary, it flows southwards, intersecting with the Gwato creeks before merging into the Benin River. The Benin River meanders for approximately 250 kilometers across Edo and Delta States in Southern Nigeria before finally draining into Atlantic Ocean (Igoni, 2018).

3.1.2 Climate

This means that the region experiences distinct wet and dry seasons throughout the year. The average annual temperature in Ovia River ranges from 25°C to 30°C. The hottest months are typically between February and April, with temperatures reaching up to 35°C. Ovia River receives the majority of its rainfall during the wet season, which typically lasts from April to October with the heaviest rainfall typically occurring in July and August. The average annual rainfall in the region is around 1,500mm to 2,000mm (Ikhile, 2018). Due to its tropical climate, Ovia River experiences high levels of humidity throughout the year (Ilaboya *et al.* 2024). Humidity levels can range from 70% to 90% during the wet season. Overall, the climate of Ovia River in Benin City, Edo State is characterized by its tropical wet and dry climate, with distinct wet and dry seasons, high humidity, and lush vegetation.

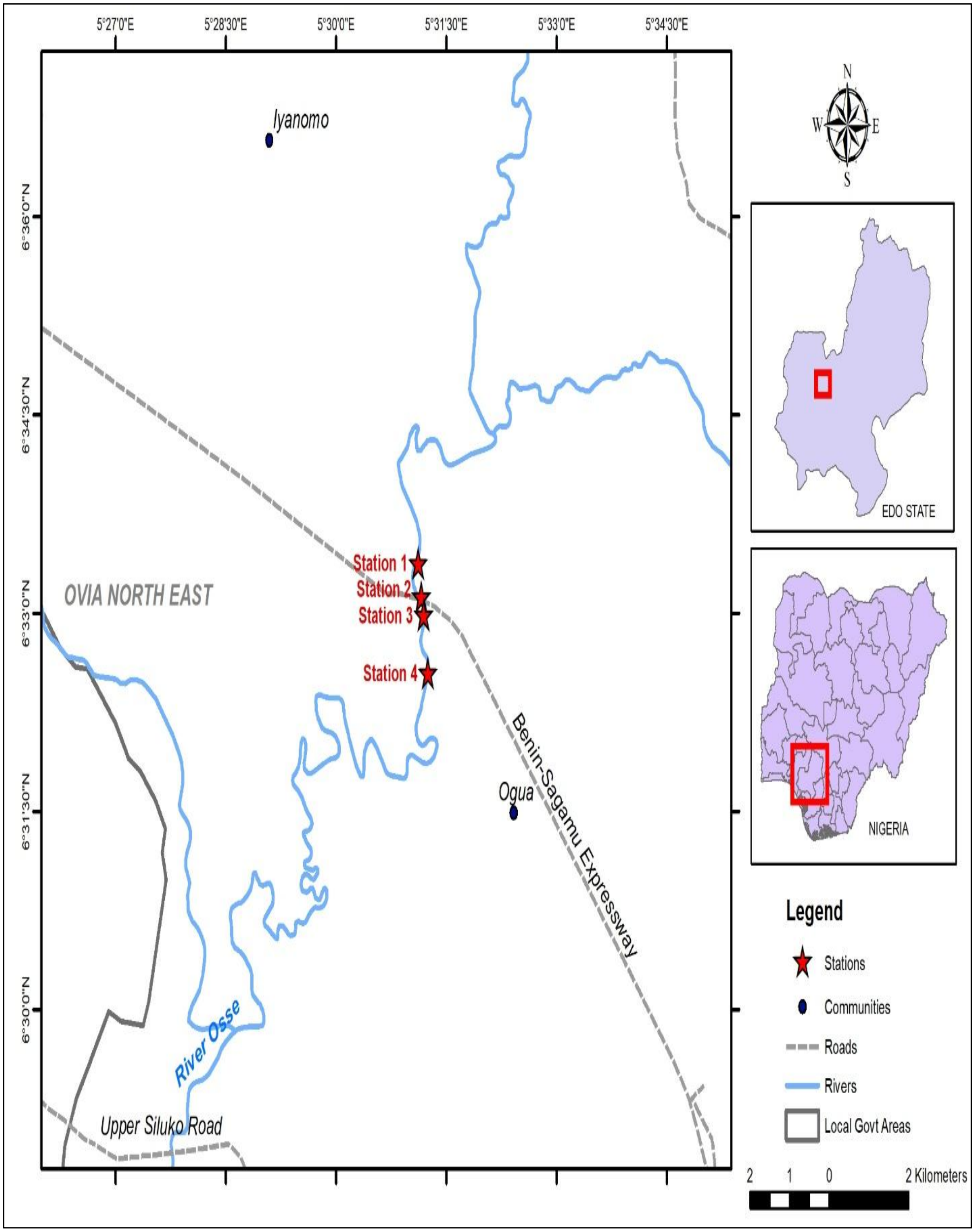

Fig. 3.1: Ovia River Showing Study Stations with: Inset 1: Map of Nigeria showing the location of the Study Area; Inset 2: Map of Edo State showing the location of the Study Area.

3.1.3 Geology

The area is characterized by sedimentary rocks such as sandstone, shale, and limestone. These rocks were formed through processes like deposition, compaction, and cementation (Oyegun and Oyegun, 2015). The formation of the Ovia River basin can be attributed to tectonic activities, erosion, and sedimentation processes over millions of years. The river has carved out a deep valley, shaping the unique landscape of the region (Oyegun and Oyegun, 2017). Hydrologically, the Ovia River receives water from various tributaries and streams, serving as a vital water source for irrigation, drinking, and industrial purposes in the surrounding areas. The river plays a crucial role in the local ecosystem and the economy (Egbai and Oyegun, 2019).

3.1.4 Vegetation and Land Use

The catchment of the study area is composed mostly of secondary rainforest which has been generally subjected to extensive deforestation and it is assumed to be due to increase in the population size of the settlement around the river. Presently, the vegetation is composed of rubber trees (*Havea brasiliensis*), Palm trees (*Elaeis guineensis*), Bambo trees (*Bambusa sp*) and shrubs. On the surface of the river, are the floating macrophytes such as water hyacinth (*Eichhornia crassipes*), water fern (*Salvinia sp*), Duckweed (*Lemna sp*) among others.

3.1.5 Sampling Station

From Upstream to downstream, four locations were chosen along the river as study stations.

Station 1: It is located at the upstream of the study area. It lies on Latitude 6.55627° N and Longitude 5.51852° E. This station is a clear water zone and the river bank is flanked by oil palm trees (*Elaeis guineensis*), rubber trees (*Havea brasiliensis*) which provide a canopy for the underlying vegetation including the antelope grass (*Echinochlo apyramidalis*), some floating macrophytes like the water lettuce (*Nymphaea lotus*). Apart from occasional fishing activities, anthropogenic disturbance at this site is negligible. Sand and organic debris in various stages of degradation are found in the surface of the ground (Plate 3.1).

Station 2: This station is about 5km downstream of station 1 (Latitude 6.55121°N and Longitude 5.51925°E). The principal anthropogenic activities observed at this site were fishing and small-scale farming around the river bank whose wastes generated are channeled directly to the river which contributes to the nutrient loading and sedimentation in river. Domestic waste is observed which may lead to localized pollution. The vegetation was sparse with no trees but some grasses, shrubs and macrophytes such as *Eichhornia crassipes* and *Nymphaea lotus* was present. The substratum is composed of mud and decaying organic materials (Plate 3.2)

Plate 3.1: Station 1

Plate 3.2: Station 2

Station 3: This station is about 5km downstream of station 2. It is located on latitude 6.54979°N and Longitude 5.51925°E. The river in this portion is surrounded on the bank by arable farmland. The crops normally cultivated include; Cassava, yam and maize. The substratum is composed of organic, rich sticky mud and fine silt (Plate 3.3). Farming, fishing and hunting constitutes the major forms of human activities at this station and the river had floating macrophytes like *Salvinia* sp, *Nymphaea* sp, *Lemna* sp and *Eichhornia crassipes* (water hyacinth).

Station 4: This is situated about 5km downstream of station 3 (latitude 6.54242°N and longitude 5.52077°E). The dominant anthropogenic activities at this site were fishing, bathing, and washing. Decomposing plants matters and small amounts of organic material are combined with clay to form the substrate. The vegetation includes tropical trees, shrubs, and aquatic plants, but there are clear signs of deforestation (Plate 3.4).

Plate 3.3: Station 3

Plate 3.4: Station 4

3.2 FIELD SAMPLING

3.2.1 Collection of Surface Water Samples

Surface water samples were collected from four designated stations along the Ovia River, covering transects from the shoreline to the open water zone. Sampling was conducted between January to June, 2024 with collections made for both physicochemical parameters and microplastic analysis.

For physicochemical analysis, surface water samples (0.5 to 1 m depth) were collected using plastic bottles, which are chemically inert and ideal for general water quality parameters such as pH, conductivity, turbidity, and nutrients. The bottles were thoroughly rinsed with deionized water prior to sampling. For dissolved oxygen (DO) analysis, water samples were collected directly into 300 mL glass-stoppered DO bottles, and immediately fixed on-site using Winkler reagents to prevent changes in DO concentration. All samples were stored in an ice-filled cooler and transported promptly to the laboratory for further analysis.

For microplastic analysis, surface water samples were also collected using 1 L glass bottles, which are suitable for avoiding plastic contamination. To ensure consistency, two replicates were taken at 5-meter intervals at each station and then combined to form a composite sample. The glass bottles were pre-rinsed with deionized water, tightly sealed after sampling, and placed in a cooler containing ice to preserve sample integrity during transport. A total of 18 composite water samples were collected across all sampling stations and months for the purpose of microplastic evaluation. All collected samples were subsequently analyzed in the laboratory following standard analytical protocols.

3.2.2 Collection of fish samples

Fish samples for MP analysis were caught from the Ovia River by local fishermen using sets of gill nets and hand nets. Often the fish arrived alive at the landing zone, so the entire sample was euthanized in ice, which also reduced digestive action and preserved stomach content. Two (2), species of commercially important fish, *Heterobranchus bidorsalis* and *Notopterus afer* were picked at each sampling. The fish were transported in coolers to the laboratory (between one and two hours travel time) where they were frozen (−20 °C) for further processing.

3.2.3 Sampling Periodicity

Water samples were collected about 10 cm below the surface water from Ovia River from January to June, 2024. The surface water samples were collected against water current and were homogenized to form a sample (composite) for that point. In advance to the sampling, all containers used for the water collection were carefully washed and labelled. This was done for all sampling points in Ovia River. Overall, six sets of samples were collected from each sampling site using clean quart glass bottles (one liter). The bottles were capped tightly, shielded from light and stored at 4°C to prevent evaporation and transported to the laboratory. Sampling was done between the hours of 6:30 am and 9 am on each sampling day. The rationale for collecting surface water was due to the ability of microplastics accumulating in surface water microlayer due to their microsize dimension. (Enyoh *et al*. 2019).

3.3 DETERMINATION OF THE PHYSIOCHEMICAL PARAMETERS OF SURFACE WATER

3.3.1 Water Temperature (°C)

Ambient and surface water temperature was determined in the field with mercury-in-glass thermometer (0-100°C). The air temperature was measured by holding the thermometer in a shaded area away from direct sunlight for a few minutes before recording the temperature. To measure the water temperature, the thermometer was gently immersed into the water, allowing it to adjust before recording the reading. The temperatures were recorded once the thermometer had fully adjusted to the specific conditions of each medium.

3.3.2 Turbidity (NTU)

The turbidity was measured using a VS72IG transparent spectrophotometer, which expresses the results in NTUs (Nephelometeric Turbidity Units). To ensure accurate readings, the cuvettes were cleaned and rinsed with distilled water. One cuvette was filled with the sample, and the other with distilled water, serving as a reference. The sample was then measured at 420 nm wavelength.

3.3.3 Electrical Conductivity (μS/cm)

A conductivity meter (HACH 44600-00) was used to measure the conductivity of water samples in $\mu \mathrm{S} cm^{-1}$. As soon as a stable reading was achieved, it was recorded after dipping the probe into the sample's container. When taking the reading, care was taken to prevent the meter's terminals from coming into touch with the beaker's wall.

3.3.4 Total Solids (TS) (mg/L)

Total solids (TS) represent the sum of both dissolved and suspended matter present in water. The parameter was determined gravimetrically according to standard procedures (APHA, 2007). A known volume of the water sample was evaporated in a pre-weighed dish and dried to a constant weight in an oven at 103–105 °C. The increase in weight of the dish after drying represents the total solids content of the sample, expressed in milligrams per liter (mg/L).

$$TS = TDS + TSS$$

3.3.5 Total Dissolved Solids (TDS) (mg/L)

The total dissolved solids were determined using the conductivity meter (HACH 44600-00). The sample's conductivity was measured using a conductivity meter, and TDS was calculated by multiplying the result by 0.666.

TDS (mg/L) = Conductivity value × 0.666 (Sawyer *et al*. 2003).

3.3.6 Total Suspended Solid (TSS) (mg/L)

This was computed by deducting the calculated amount of the TDS from the TS.

$$TSS = TS - TDS$$

3.3.7 Hydrogen Ion Concentration (pH)

The pH of the water samples collected was determined in the laboratory using the Hanna pH meter instrument Hi-1922 model. The meter was calibrated with pH buffer solution of 4.0, 7.0, 9.0, rinsed severally with distilled water. A 20 ml of the sample from each station was measured into beakers that were first rinsed with distilled water and then small quality of the water sample.

The pH meter probe was then immersed in the beaker containing the sample until the reading from the digital display was stable and recorded.

3.3.8 Hardness

The titrimetric method employing EDTA (Ethylene diamine tetraacetic acid) with Eriochonl, was utilized to determine the total hardness. In this method, a water sample was first treated with a buffer solution to maintain a constant pH. Eriochonl indicator was then added to the solution, imparting a characteristic color change at the endpoint of the titration. EDTA titrant was incrementally added to the sample until the color transition was observed, indicating the completion of the reaction. The volume of EDTA titrant used in the titration was then used to calculate the total hardness of the water sample, typically expressed in mg/L or ppm of calcium carbonate ($CaCO_3$) equivalent.

3.3.9 Alkalinity

To determine the alkalinity, 4 drops of phenolphthalein were introduced into a 100 ml water sample. If the color shifted to red, the sample was titrated with 0.01 N H_2 SO_4. The endpoint value was noted when the red precipitate vanishes. If there was no color change post-phenolphthalein addition, the alkalinity was zero. Subsequently, the total alkalinity test was conducted by adding 3 drops of methyl orange indicator to the water sample. Titration against 0.05 N H_2 SO_4 from a burette continued until the color transitioned from yellow to orange. The endpoint value was recorded in mg/L. Alkalinity (mg CaCO_3 ^{3} L^{-1}) is calculated using the formula;

$$\text{TA} = \frac{1000 \times VA \times M}{100}$$

Where VA = volume of acid used, and

M = Mass of CaCO_3 equivalent to 1ml of titrant (5.00mgl^{-1} for 0.05m H_2SO_4) (APHA, 2012).

3.3.10 Dissolved Oxygen (mgL^{-1})

It was determined using the Winkler method (APHA, 1998). In the field, samples were taken in 250 ml DO glass bottles with stoppers. To prevent air bubbles from getting into the water samples, the bottles were filled and covered below the water's surface. The sample was treated with 1.5 ml each of Winkler's solutions A (Manganous Sulphate, MnSO_4) and B (Potassium iodide in phosphate hydroxide), resulting in a brown precipitate formation indicating the trapped oxygen. In the laboratory, to dissolve the precipitate, two (2) ml of concentrated sulfuric acid (H_2SO_4) were applied to the sample. A 250 ml conical flask was filled with an aliquot of 100ml, two drops of freshly made starch Indicator, and was vigorously stirred. To make the solution colourless, it was titrated against 0.025 M sodium thiosulphate solution ($Na_2S_2O_3.5H_2O$). The quantity of sodium thiosulphate employed corresponds to the sample's DO content in mg/l.

3.3.11 Biological Oxygen Demand (BOD$_5$) (mgL^{-1})

The laboratory conducted an assessment of the water's biochemical oxygen demand. Water samples were collected using 250 ml reagent glass bottles, with stoppers replaced beneath the water surface to prevent the entry of atmospheric oxygen and the bottles secured in black polythene bags to stop the further effect of photosynthesis which leads to the production of more oxygen in the collected sample. Biochemical Oxygen Demand involves quantifying the oxygen needed to oxidize the organic substances in the sample over a 5-day incubation period at 20°C.

Dissolved oxygen levels were measured at the start and end of the incubation, and BOD was determined by subtracting the initial DO value from the final DO value (APHA, 2012).

$$BOD_5 \text{ (mg/l)} = DO_1 - DO_5$$

Where DO_1 = Dissolved oxygen at the beginning of the incubation period (initial DO)

DO_5 = Dissolved oxygen at the end of the incubation period (final DO)

3.3.12 Chemical Oxygen Demand (COD) (mgL^{-1})

The COD of the samples was determined using the dichromate method, which involved pipetting 50 ml of the sample into a conical flask along with 10 ml of 0.00833 $K_2CR_2O_7$ solution, 1 ml of H_2SO_4 and 10 ml of $AgSO_4$ before gently heating to boiling for precises 10 minutes on a hot plate with a plastic funnel on the conical flask's mouth. 30 minutes were spent cooling the mixture. Following that, 2 drops of Ferron indicator were added, and the colour were tested against 0.02M $Fe(NH_4)2.6H_2O$ until it turned red brown instead of blue-green. To determine the actual the value of COD, the COD of a predefined blank was deducted from that of the sample.

3.3.13 Exchangeable Bases (Sodium, Potassium, Calcium and Magnesium)

Flame photometric method was implemented in the determination of the exchangeable bases (sodium, potassium, calcium and magnesium) concentration in the surface water samples. This was ascertained employing the Technicon auto analyzer flame photometer IV, which was internally calibrated using known amount of lithium and the elements Na, K, Ca, and Mg. Samples were placed in identical cups in the sample tray module and instantaneously sucked into the mixing module, where the lithium and the sample were mixed. The Teflon tube was periodically checked for a satisfactory bubble pattern. After that, the combined sample were introduced into the flame chamber, where it was heated by propane gas, atomized and flared. Each anion's concentration was determined by measuring the flame's colour intensity, which was recorded on an affixed recorder.

3.3.14 Chloride (Cl^-)

Using MOHR's methodology, this was determined (APHA, 2005). This approach uses $K_2CR_2O_4$ as the end point indicator and silver nitrate ($AgSO_3$) as the titrant. The sample's chloride ion precipitates out as white sliver chloride. First, a reddish-brown colour comparison blank was made by mixing 100 ml of distilled water with 1ml of potassium chromate and 0.2 ml of 0.02 M $AgSO_3$ in a clean conical flask. After a little shake, it was allowed to stand. Then, 100 ml of the sample was pipetted into a conical flask together with 1 ml of the potassium chromate Indicator, and 0.02 M $AgSO_3$ was titrated while stirring continuously until a faint red precipitate is formed. The value of chloride ion concentration was extrapolated from the equation;

$$\text{Cl- (mg/l)} = \frac{Molarity \times Titre \times Mol.Wt}{Aliquot\ taken}$$

3.3.15 Sulphate (SO_4^{2-})

This was determined by the colorimetric technique where 5 ml of sample was pipetted into a 50 ml flask, 1 ml of Gelatin-$BaCl^{2-}$ reagent was added and 14 ml of distilled water was added to make it up to 25 ml, the solution was well shaken in between reagent addition. The absorbance was then read at 420 nm using the visible spectrophotometer VS72IG.

$$SO_4\ \text{(mg/l)} = \frac{Instrument\ reading \times Slope\ reciprocal \times Col.vol}{Aliquot\ Taken}$$

3.3.16 Phosphate

This was determined by the ascorbic acid method (APHA, 2005). Ascorbic acid, antimony potassium tartrate, sulfuric acid and ammonium molybdate are employed in this process as a combination reagent. A 50 ml conical flask was filled with 2.5 ml of the mixed reagents, 5 ml of the sample, and 98% ethanol. Before reading absorbance at 880 nm using the spectrophotometer VS72IG, absorbance was given 30 minutes to develop into a blue colour.

$$\text{PO4 (mg/l)} = \frac{Instrument\ reading \times Slope\ reciprocal \times Col.vol}{Aliquot\ Taken}$$

3.3.17 Nitrate

Nitrate levels were determined colorimetrically using the Metton Roy Spectronic 21D spectrometer. The concentrations of unknown samples were derived from a standard curve and recorded in mg/l. The Brucine method was employed for nitrate analysis. A 10 ml portion of the water sample was placed in a Nessler tube, and another Nessler tube received 10 ml of distilled water for use as a blank. To both tubes, 10ml of Brucine was added. Subsequently, 20 ml of $H_2\mathrm{S}O_4$was introduced to the water sample. The water sample was titrated with $\mathrm{KN}O_3$ until its color matched that of the blank. The nitrate concentration in mg/l was then calculated using the formula provided (APHA, 2012).

$$\mathrm{NO_3\ (mg/l)} = \frac{Volume\ of\ \mathrm{KN}O_3 \times 1000}{Volume\ of\ sample}$$

3.3.18 Heavy Metals

Water samples were digested using aluminum block digester 110. Digestion of the water sample was done by introducing 4ml of perchloric acid, 20 ml of concentrated nitric acid, and 2 ml of concentrated tetraoxosulphate VI acid to 100 ml of the sample. Aluminum block digester 110 was used to break down this. Heat was applied to the mixture till white fumes and a transparent solution emerged. The samples were then transferred to 100 ml volumetric flask, given some time to cool, and then filled to the proper level with distilled water and properly mixed. The sample was let to stand overnight to separate the insoluble components and thereafter filtered using a 0.45 Milipore type filter was used to filter it.

Heavy Metals (As, Cd, Cr, Cu, Fe, Mn, Ni, PB, V, Zn) were analyzed using the Atomic Absorption spectrophotometer (AAS) Solar 969 Unicam Series model. Each metal has a hollow cathode lamp for its determination. Each element to be analyzed has its own wavelength set on the device. Between each reading, distilled deionized water was aspirated. By keeping an eye on the steady galvanometer reading every 1-2 minutes, reading of the absorbance were obtained. Triplicate analysis for each sample was done and the standard calibration plot was used to determine the metal content.

3.4 DETERMINATION OF MICROPLASTICS IN WATER SAMPLES

3.4.1 Processing and Extraction of Microplastics in Surface Water

Surface water samples as shown in Plate 4.5, were filtered through a stacked arrangement of 5.6-mm (No. 3.5) and 0.3-mm (No. 50) stainless steel mesh sieves. The solid collected were rinsed in a beaker with distilled water and placed in a 90°C drying oven for 24 hours or longer to sample dryness. The sieved material was dried to determine the solids mass in the sample. The solids were subjected to wet peroxide oxidation (WPO) in the presence of a Fe(II) catalyst to digest labile organic matter. Then, followed by the addition of 20 mL of 30% hydrogen peroxide and mixed for 5 minutes at room temperature. A stir bar was added to the beaker and heated on a hotplate at 75°C with watch glass covered on it. The beaker was removed from the hotplate as soon as gas bubbles were observed and placed it in the fume hood until the boiling subsides. About 20 ml of 30% hydrogen peroxide was added whenever the natural organic materials were visible. The WPO mixture was subjected to density separation in NaCl (aq) to isolate the plastic debris through flotation. The floating solids were separated from the denser undigested mineral components using a density separator. The floating plastic debris were collected in the density separator using a custom 0.3 mm filter, air-dried, and plastic material was removed and weighed to determine the microplastics concentration.

3.5 Determination of Microplastics in Fish

3.5.1 Processing and Extraction of Microplastics in Fish

Each fish specimen as shown in Plates 4.6-4.7, were dissected, and the entire gastrointestinal (GI) tracts were extracted by making a precise incision just below the throat and delicately cutting along the ventral side of the fish to the anal opening, separating the stomachs. Subsequently, the stomach contents were rinsed with distilled water and diluted where necessary due to an abundance of content. The GI tract (including the oesophagus, stomach, and intestine) was meticulously isolated from other tissues and placed in a container filled with a 10% KOH solution (Analytical reagent grade, Fisher Chemical). The containers were then left at room temperature for 2-3 weeks until complete dissolution of the organic material was observed. To prevent damage to plastic particles from hard stomach constituents (e.g., shells), the containers were not agitated. Once the organic material was degraded, a suction pump was used to extract the non-digestible residue during the filtration process. The residue was then dried at a controlled temperature of 30 °C to preserve the integrity of the microplastics.

Plate 3.5: Surface water samples used for Microplastics Analysis

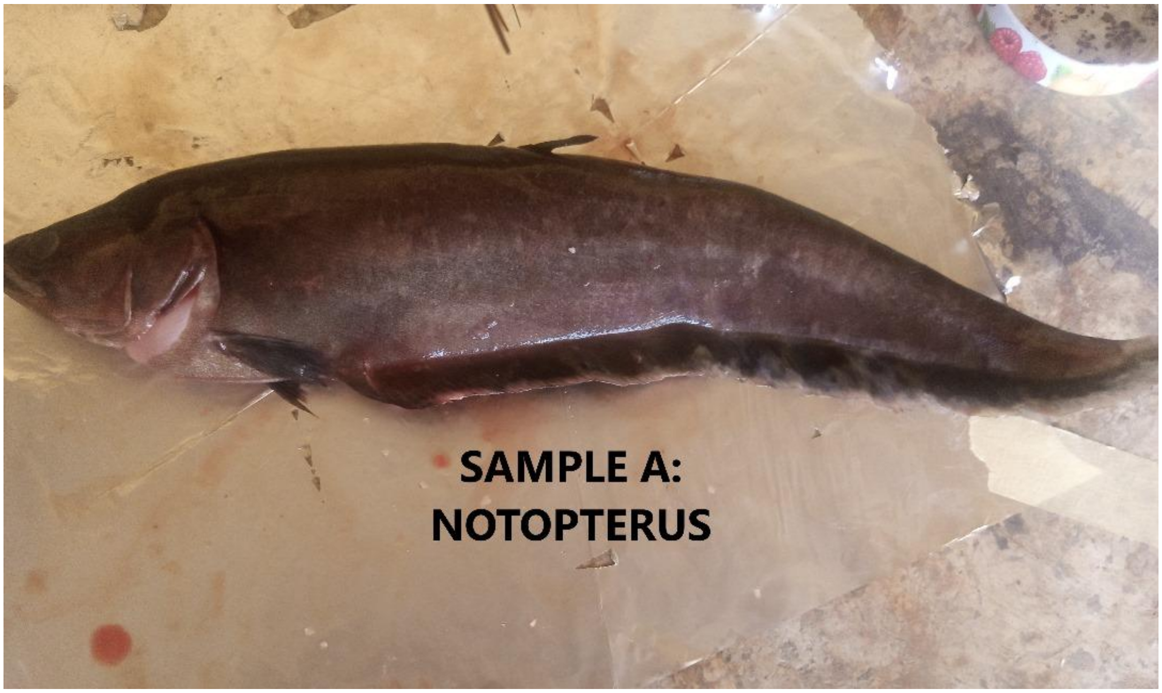

Plate 3.6**:** *Notopterus afer*

Plate 3.7: *Heterobranchus bidorsalis*

3.6 Identification, Characteristization and Quantification of Microplastics

All microplastic samples were carefully transferred to sample carriers for analysis. Fourier Transform Infrared (FTIR) spectroscopy (Nicolet iS10 FTIR spectrometer) was employed to identify the functional groups and chemical properties of the polymers. The samples were analyzed using Attenuated Total Reflection (ATR) mode within the mid-infrared range of 650–4000 cm^{-1}, with 18 scans per analysis to ensure spectral accuracy. To examine the size, shape, and surface characteristics of microplastic particles, samples underwent ultrasonic cleaning before being visualized using Scanning Electron Microscopy (SEM) combined with Energy Dispersive X-ray Spectroscopy (EDS). SEM provided high-resolution images of the microplastics, which were then processed in ImageJ software for quantitative analysis. ImageJ facilitated the measurement of particle dimensions and morphology, enabling statistical characterization of the microplastic size distribution. For the particle size distribution (PSD) data, a quality control criterion was applied: results were accepted only if the mean particle size was greater than the standard deviation (Mean > SD). If the mean was less than the standard deviation, the PSD result was considered unreliable and thus rejected. Qualitative elemental composition of the microplastic particles was confirmed using EDS (X-Act, Oxford), which provided atomic percentage data of key elements associated with the microplastics.

3.7 Water Quality Index (WQI)

The portability of water sources has been defined using water quality index (WQI) which provides a single number or grade that expresses overall water quality at a certain location and time, based on the composite influence of a number of water quality parameters. (Shweta *et al.* 2013).

The water quality index (WQI) was calculated using weighted arithmetic water quality index method. In this method, different water quality components were multiplied by a weighting factor and were then aggregated using simple arithmetic mean. (Shweta *et al.* 2013; Oboh and Agbala 2017; Egun and Ogiesoba-Eguakun 2018).

The Water Quality Index (WQI) was calculated using the following equation:

$$WQI = \Sigma W_i Q_i / \Sigma W_i$$

The quality rating scale (Qi) for each parameter was calculated using

$$Q_i = 100 [(V_i - V_0) / (S_i - V_0)]$$

Where:

Vi = estimated concentration of the ith parameter of interest in the analyzed water.

V0 = ideal value of the ith parameter in pure water

V0 = 0 except for pH=7 and DO=14.6mgl^{-1}

Si = Recommended standard value of ith parameter

The unit weight (Wi) for each water quality parameter is calculated by using the following formula: $W_i = K / S_i$

Where K = proportionality constant and can also be calculated using the following equation:

$K = 1 / \Sigma (1 / S_i)$

The rating for water quality index is given in Table 3.1

3.8 Comprehensive Pollution Index (CPI)

Comprehensive Pollution Index (CPI) is the arithmetic mean of the number of chosen pollutants in the water. The Comprehensive Pollution Index (CPI) is calculated using the following equation:

$$\mathrm{CPI} = 1/n \sum_{i=1}^{n} ci / si$$

Where, C_i is the measured concentration of the pollutant (mg L^{-1}),

S_i is the National water quality standard permissible limit for the pollutant in surface water,

n is the number of chosen pollutants. (Egun and Oboh 2021, Li *et al*. 2010)

CPI is classified into the water quality levels listed in Table 3.2

Table 3.1 Water quality rating according to weighted arithmetic water quality index method

Levels	Rating of water quality	Grading
0-25	Excellent water quality	A
25-50	Good water quality	B
50-75	Poor water quality	C
76-100	Very poor water quality	D
>100	Unsuitable for drinking purposes	E

Table 3.2 Standard surface water quality categories based on CPI (Li *et al*. 2010)

The Comprehensive Pollution Index (CPI)	Level	Explanation of the water quality grades
Less than 0.2	I	Cleanness
0.21–0.4	II	Sub-cleanness
0.41–1.0	III	Slight pollution
1.01–2.0	IV	Moderate pollution
More than 2.01	V	Severe pollution

3.9 Risk Assessment

3.9.1 Chronic daily intake (CDI)

The Chronic Daily Intake (CDI) represents the estimated daily dose of contaminants that an individual is exposed to over a prolonged period. CDI is calculated using the following equation:

$$\text{CDI}=\frac{C \times IR \times EF \times ED}{BW \times AT}$$

where:

- **C** = Concentration of the contaminant in the medium (e.g., µg/l for water)
- **IR** = Ingestion rate (L/day)
- **EF** = Exposure frequency (days/year)
- **ED** = Exposure duration (years)
- **BW** = Body weight (kg)
- **AT** = Averaging time (days) – for non-carcinogenic risk, this is typically ED × 365, and for carcinogenic risk, it is often taken as a lifetime (e.g., 70 years × 365 days)

Determination of Parameters

- Ingestion rate (IR): The rate of water intake was based on standard defaults. For adults, 2.0 L/day is widely applied (USEPA, 2011; ATSDR, 2018), though more conservative estimates use 2.4 L/day. WHO guideline derivations often apply 2.0 L/day with an assumed body weight of 60 kg. In this study, an adult ingestion rate of 2.0 L/day was adopted.

- Exposure frequency (EF): EF represents the number of days per year an individual is exposed. While some studies assume daily intake (365 days/year), a frequency of 350 days/year is commonly recommended to account for time spent away from the exposure source. In this study, 350 days/year was applied.
- Exposure duration (ED): This reflects the length of exposure in years. Typical values include 6 years for children and 30 years for adults under residential exposure scenarios. An exposure duration of 30 years was selected for this assessment.
- Body weight (BW): Standard assumptions vary with guidance documents. Historically, 70 kg has been used as the mean adult body weight, though updated references suggest 80 kg (USEPA, 2011). WHO often applies 60 kg for guideline values. This study adopted 70 kg as the representative adult body weight.
- Averaging time (AT): For non-carcinogenic effects, AT = ED × 365 days. For carcinogenic effects, AT is taken as a lifetime, typically 70 years × 365 days = 25,550 days (USEPA, 2011).

Accordingly, the parameter values used for this study were: IR = 2.0 L/day, EF = 350 days/year, ED = 30 years, BW = 70 kg, and AT = ED × 365 days (non-carcinogenic) or 25,550 days (carcinogenic). These values follow widely applied defaults in health risk assessment (USEPA, 2011; WHO, 2017).

3.9.2 Carcinogenic and Non-Carcinogenic Risk Assessment

Once the CDI was estimated, the potential health risks were evaluated by calculating the Carcinogenic Risk (CR) and the Hazard Quotient (HQ) for non-carcinogenic effects.

Carcinogenic Risk (CR):

Carcinogenic Risk (CR) represents the probability of an individual developing cancer over a lifetime as a result of exposure to a carcinogenic contaminant. It is calculated using the following equation:

$$CR = CDI \times CSF$$

Where:

- CDI = Chronic Daily Intake of the contaminant (mg/kg-day)
- CSF = Chemical-specific Carcinogenic Slope Factor (mg/kg-day^{-1})

The CSF is a toxicity value that quantifies the increased lifetime cancer risk per unit of contaminant intake. Each heavy metal has a specific slope factor derived from toxicological data and published by agencies such as the U.S. Environmental Protection Agency (EPA) Integrated Risk Information System (IRIS) or related health agencies. For example, arsenic has a CSF of 1.5 (mg/kg-day^{-1}), chromium (VI) has 0.5 (mg/kg-day^{-1}), while some metals such as cadmium and lead do not have oral slope factors available in IRIS. In such cases, carcinogenic risk estimation was not applied. This approach ensures that the cancer risk assessment reflects the unique carcinogenic properties of each contaminant studied.

A CR value greater than 1×10^{-6} was considered to be of concern, although acceptable risk levels may vary based on regulatory guidelines.

Non-Carcinogenic Risk (HQ):

The non-carcinogenic risk was evaluated using the Hazard Quotient (HQ), which represents the ratio of the estimated exposure (CDI) to the contaminant's reference dose (RfD):

$$HQ = \frac{CDI}{RFD}$$

Where:

- CDI = Chronic Daily Intake (mg/kg-day)
- RfD = Reference Dose (mg/kg-day)

The RfD is the maximum acceptable oral dose of a toxic substance that is not expected to cause adverse health effects during a lifetime of exposure. It is specific to each chemical and derived from toxicological studies, usually incorporating uncertainty (safety) factors.

An HQ value below 1 suggests that the exposure is unlikely to cause adverse health effects, whereas an HQ above 1 indicates a potential health risk.

3.10 Data Analysis

Two-factor analysis of variance (ANOVA) was used to test if there was a significant difference in average microplastic contamination levels across the four stations, over time which is crucial for understanding the spatial and temporal distribution of microplastics in the study area. Additionally, ANOVA was also applied to physiochemical parameters, utilizing basic statistical methods of central tendency and dispersion to describe and compare characteristics across different sampling stations. The computer programming SPSS 16.0 and Microsoft Excel (2007) for Windows were used to perform the Duncan Multiple Range (DMR) test to locate the significance difference if $P < 0.05$.

CHAPTER FOUR

RESULTS

4.1 Physical and Chemical Parameters of Surface Water

The results of the physical and chemical characteristics of surface water from the study stations along the stretch of Ovia River are summarized in Table 4.1. The mean, standard deviation, minimum and maximum values for each parameter analyzed at the four stations are given. Also shown are the p-Values of the one-way analysis of variance (ANOVA).

In Table 4.1, the following physical and chemical properties such as temperature, pH, electrical conductivity, turbidity, total suspended solids, total dissolved solids, dissolved oxygen, biochemical oxygen demand, chemical oxygen demand, alkalinity, sodium, magnesium, potassium, calcium, hardness, chloride, nitrate, sulphate, phosphate, cadmium, chromium, iron, zinc and nickel showed no significant difference ($p > 0.05$) while manganese, lead and copper exhibited a significant difference ($p < 0.05$) across the study stations in Ovia River. Arsenic and vanadium were below detection limits.

4.1.1 Water Temperature (°C)

The spatial and temporal variations in temperature are shown in Fig. 4.1. Spatially, station 1 recorded the highest mean temperature of 28.57 °C, followed by station 2 with 28.40 °C, station 3 with 28.37 °C, and station 4 with the lowest average of 28.30 °C. Temporally, the mean temperature was highest in March at 29.88 °C, January recorded a mean of 27.70 °C, while May had the lowest average temperature of 27.65 °C. ANOVA (Analysis of Variance) indicates no significant differences in temperature among the study stations ($P > 0.05$).

Table 4.1: Summary of the Physical and Chemical Parameters of Surface Water of Ovia River (January-June, 2024)

Parameter	Station 1 Mean ± SD (Min - Max)	Station 2 Mean ± SD, (Min - Max)	Station 3 Mean ± SD (Min - Max)	Station 4 Mean ± SD, (Min - Max)	Pvalue	WHO Limits	USEPA Limits	FMEnv Limits
Temperature (°C)	28.57±1.33 (27.70 - 30.10)	28.40±1.21 (27.70 - 29.80)	28.37±1.24 (27.60 - 29.80)	28.30±1.30 (27.50 - 29.80)	P>0.05	15 - 35		25 - 30
pH	6.73±0.21 (6.50 - 6.90)	6.73 ± 0.06 (6.70 - 6.80)	6.87 ± 0.25 (6.60 - 7.10)	6.87 ± 0.21 (6.70 - 7.10)	P>0.05	6.5 - 8.5	6 – 9	6.5 - 8.5
EC (µS/cm)	83.00±33.87 (56.00 -121.00)	79.33 ± 31.34 (53.00 -114.00)	81.33 ± 33.61 (52.00 - 118.00)	81.00 ± 31.76 (54.00 - 116.00)	P>0.05	< 2,500	< 2,000	< 1000
Turbidity (NTU)	8.32 ± 2.90 (5.16 - 10.85)	6.06 ± 3.81 (2.15 - 9.76)	8.49 ± 2.41 (6.24 - 11.03)	7.09 ± 2.64 (4.13 - 9.20)	P>0.05	< 5	< 5	< 5
TSS (mg/L)	3.30 ± 1.85 (1.50 - 5.20)	2.60 ± 1.44 (1.00 - 3.80)	3.20 ± 0.75 (2.50 - 4.00)	2.77 ± 1.23 (1.40 - 3.80)	P>0.05	< 30	0 - 160	< 25
TDS (mg/L)	41.33 ± 16.65 (28.00 - 60.00)	39.00 ± 15.39 (26.00 - 56.00)	40.00 ± 15.72 (26.00 - 57.00)	40.33 ± 15.95 (27.00 - 58.00)	P>0.05	< 500	< 500	< 500
DO (mg/L)	6.53 ± 1.22 (5.20 - 7.60)	6.60 ± 0.72 (5.80 - 7.20)	6.33 ± 0.90 (5.40 - 7.20)	6.67 ± 1.21 (5.40 - 7.80)	P>0.05	> 5	> 4	> 5
BOD (mg/L)	3.60 ± 0.53 (3.20 - 4.20)	3.47 ± 0.42 (3.00 - 3.80)	3.33 ± 0.31 (3.00 - 4.00)	3.53 ± 0.23 (3.40 - 3.80)	P>0.05	< 5	< 5	< 5
COD (mg/L)	26.00 ± 5.29 (22.00 - 32.00)	20.33 ± 3.21 (18.00 - 24.00)	23.33 ± 8.33 (14.00 - 30.00)	22.67 ± 5.03 (18.00 - 28.00)	P>0.05	< 25	< 40	< 25

Parameter	**Station 1** **Mean ± SD** **(Min - Max)**	**Station 2** **Mean ± SD,** **(Min - Max)**	**Station 3** **Mean ± SD** **(Min - Max)**	**Station 4** **Mean ± SD,** **(Min - Max)**	**Pvalue**	**WHO Limits**	**USEPA Limits**	**FMEnv Limits**
Alkalinity (mg/L)	15.83 ± 3.82 (12.50 - 20.00)	13.75 ± 3.31 (11.25 - 17.50)	15.83 ± 5.20 (10.00 - 20.00)	14.17 ± 1.44 (12.50 - 15.00)	P>0.05	20 - 120	20 - 200	20 – 100
Sodium (mg/L)	5.35 ± 1.63 (3.85 - 7.09)	5.07 ± 1.53 (3.72 - 6.73)	5.30 ± 1.57 (3.69 - 6.82)	5.56 ± 1.61 (3.73 - 6.78)	P>0.05	< 200	< 200	< 200
Magnesium (mg/L)	2.85 ± 0.96 (1.75 - 3.50)	2.59 ± 1.47 (1.56 - 4.28)	2.66 ± 1.17 (1.56 - 3.89)	2.92 ± 1.01 (1.75 - 3.50)	P>0.05	< 30		< 25
Potassium (mg/L)	3.72 ± 1.08 (2.96 - 4.95)	3.49 ± 0.92 (2.88 - 4.55)	3.66 ± 1.07 (2.74 - 4.83)	3.54 ± 0.96 (2.80 - 4.63)	P>0.05	< 200	< 200	< 200
Calcium (mg/L)	5.88 ± 3.34 (3.21 - 9.62)	5.88 ± 3.05 (2.89 - 8.98)	5.35 ± 2.89 (2.57 - 8.34)	5.45 ± 2.74 (2.89 - 8.34)	P>0.05	< 75		< 30
Hardness (mg/L)	26.42 ± 11.63 (15.21 - 38.43)	25.35 ± 13.45 (13.61 - 40.03)	24.29 ± 12.05 (12.81 - 36.83)	25.62 ± 10.50 (14.41 - 35.23)	P>0.05	< 300		< 200
Chloride (mg/L)	23.15 ± 9.91 (15.95 - 34.45)	20.09 ± 7.38 (14.18 - 28.36)	22.15 ± 8.45 (14.18 - 31.01)	20.68 ± 6.88 (15.07 - 28.36)	P>0.05	< 250	< 250	< 250
Nitrate (mg/L)	2.67 ± 1.03 (1.49 - 3.38)	2.53 ± 0.79 (1.62 - 2.99)	2.68 ± 1.33 (1.49 - 4.12)	3.03 ± 0.56 (2.71 - 3.68)	P>0.05	< 10	< 10	< 10
Sulphate (mg/L)	1.74 ± 0.31 (1.53 - 2.09)	1.39 ± 0.42 (1.00 - 1.83)	1.50 ± 0.59 (1.06 - 2.17)	2.00 ± 0.57 (1.46 - 2.59)	P>0.05	< 250	< 250	< 150
Phosphate (mg/L)	0.14 ± 0.05 (0.08 - 0.17)	0.13 ± 0.03 (0.10 - 0.15)	0.14 ± 0.04 (0.10 - 0.18)	0.17 ± 0.04 (0.13 - 0.21)	P>0.05	< 0.1	< 1	< 0.1

Parameter	**Station 1**	**Station 2**	**Station 3**	**Station 4**	**Pvalue**	**WHO Limits**	**USEPA Limits**	**FMEnv Limits**
	Mean ± SD (Min - Max)	**Mean ± SD, (Min - Max)**	**Mean ± SD (Min - Max)**	**Mean ± SD, (Min - Max)**				
Arsenic (mg/L)	BDL	BDL	BDL	BDL		< 0.01	< 0.01	< 0.01
Cadmium (mg/L)	0.00 ± 0.00 (0.00 - 0.00)	0.00 ± 0.00 (0.00 - 0.00)	0.00 ± 0.01 (0.00 - 0.01)	0.00 ± 0.00 (0.00 - 0.00)	$P>0.05$	< 0.003	< 0.005	< 0.003
Chromium (mg/L)	0.03 ± 0.01 (0.02 - 0.04)	0.01 ± 0.01 (0.00 - 0.01)	0.03 ± 0.01 (0.02 - 0.03)	0.01 ± 0.01 (0.00 - 0.02)	$P>0.05$	< 0.05	< 0.1	< 0.05
Copper (mg/L)	0.01 ± 0.01^{a} (0.00 - 0.02)	0.02 ± 0.01^{a} (0.01 - 0.02)	0.04 ± 0.01^{b} (0.03 - 0.04)	0.01 ± 0.01^{a} (0.00 - 0.02)	$P<0.05$	< 0.2	< 1.3	< 1
Iron (mg/L)	0.14 ± 0.04 (0.11 - 0.19)	0.15 ± 0.05 (0.09 - 0.18)	0.26 ± 1.28 (0.12 - 0.37)	0.20 ± 0.8 (0.13 - 0.28)	$P>0.05$	< 0.3	< 0.3	< 0.3
Manganese (mg/L)	0.02 ± 0.01^{a} (0.02 - 0.03)	0.02 ± 0.01^{a} (0.01 - 0.01)	0.04 ± 0.01^{b} (0.03 - 0.05)	0.02 ± 0.01^{a} (0.01 - 0.02)	$P<0.05$	< 0.1	< 0.1	< 0.1
Nickel (mg/L)	0.01 ± 0.01 (0.00 - 0.02)	0.01 ± 0.00 (0.01 - 0.01)	0.02 ± 0.00 (0.02 - 0.02)	0.01 ± 0.01 (0.00 - 0.02)	$P>0.05$	< 0.02	< 0.1	< 0.02
Lead (mg/L)	0.01 ± 0.01^{ab} (0.00 - 0.02)	0.00 ± 0.00^{a} (0.00 - 0.00)	0.01 ± 0.01^{b} (0.01 - 0.02)	0.00 ± 0.00^{a} (0.00 - 0.00)	$P<0.05$	< 0.01	< 0.01	< 0.01
Vanadium (mg/L)	BDL	BDL	BDL	BDL		< 0.1	< 0.1	< 0.1
Zinc (mg/L)	0.08 ± 0.03 (0.05 - 0.10)	0.07 ± 0.01 (0.06 - 0.08)	0.12 ± 0.02 (0.09 - 0.12)	0.06 ± 0.02 (0.04 - 0.08)	$P>0.05$	< 5	< 5	< 5

Note: x ± SD = average mean generated from values across the months per station, ± standard deviation; min–max = minimum and maximum values for each parameter per station; post hoc = values with different superscripts ($a > b > c > d$) are significantly different ($p < 0.05$) while values with same superscript are not significantly different ($p > 0.05$). *$p < 0.05$ (significant difference).

4.1.2 Hydrogen Ion Concentration (pH)

The spatial and temporal variation in Hydrogen Ion Concentration (pH) measurements is shown in Fig. 4.2. Spatially, both station 1 and station 2 recorded a mean pH of 6.73, with station 1 ranging from 6.50 to 6.90 and station 2 exhibiting a narrower range of 6.70 to 6.80. Stations 3 and 4 displayed slightly higher mean pH values of 6.87, with ranges from 6.60 to 7.10 and 6.70 to 7.10, respectively. Temporally, the lowest mean pH was observed in March at 6.63, followed by January with 6.80, while May recorded the highest mean pH of 6.98. ANOVA (Analysis of Variance) revealed no significant differences in pH among the stations ($P > 0.05$).

4.1.3 Electrical Conductivity (µS/cm)

The spatial and temporal variations in electrical conductivity (EC) are shown in Fig. 4.3. Station 1 recorded the highest mean EC at 83.00 µS/cm, with values ranging from 56.00 to 121.00 µS/cm. Station 2 followed with a mean of 79.33 µS/cm, ranging from 53.00 to 114.00 µS/cm. Station 3 had a mean EC of 81.33 µS/cm, with a range of 52.00 to 118.00 µS/cm, while station 4 recorded an average of 81.00 µS/cm, ranging from 54.00 to 116.00 µS/cm. Temporally, EC values showed a clear increasing trend across the months. The lowest mean of 53.75 µS/cm was recorded in January (dry season), followed by 72.50 µS/cm in March (transition period), while May (rainy season) had the highest mean of 117.25 µS/cm. ANOVA (Analysis of Variance) indicates no significant differences in conductivity among the study stations ($P > 0.05$).

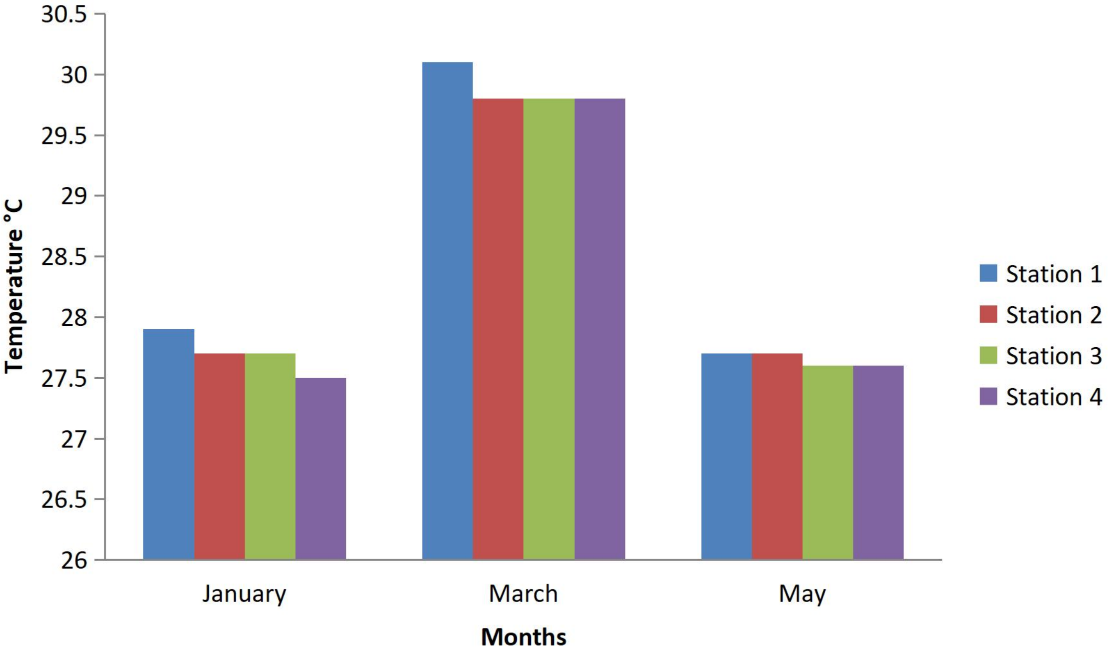

Figure 4.1: Spatial and temporal variations in Temperature (°C) at the study stations of Ovia River, Benin City

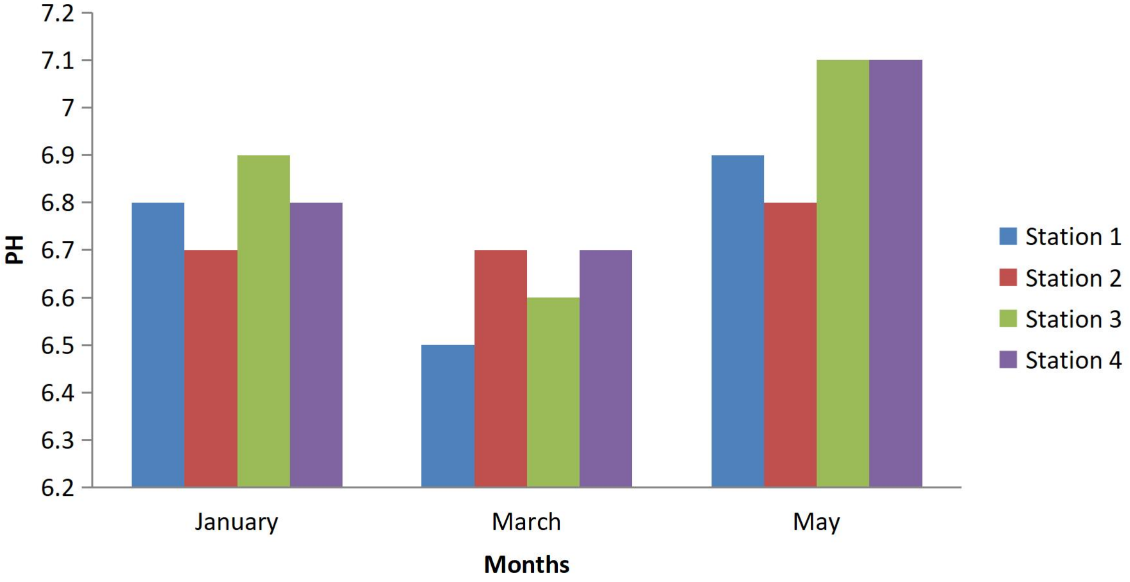

Figure 4.2: Spatial and temporal variations in pH at the study stations of Ovia River, Benin City

4.1.4 Total Suspended Solids (mg/L)

The spatial and temporary variations in total Suspended Solids measurements are shown in Fig. 4.4. Station 1 recorded the highest mean concentration (3.30 mg/L), followed by station 3 (3.20 mg/L), station 4 (2.77 mg/L), and station 2 with the lowest (2.60 mg/L). Temporally, TSS values increased steadily from January to May, rising from 1.60 mg/L in January to 3.10 mg/L in March, and reaching 4.20 mg/L in May. ANOVA showed no significant differences among the stations ($P > 0.05$).

4.1.5 Total Dissolved Solids (mg/L)

The spatial and temporary variations in the TDS are shown in Fig. 4.5. Spatially, station 1 recorded the highest mean concentration of 41.33 mg/L, followed closely by stations 4 and 3 at 40.33 mg/L and 40.00 mg/L, respectively, while station 2 had the lowest mean of 39.00 mg/L. Temporally, the mean TDS increased across sampling months, with the lowest average recorded in January at 26.75 mg/L, rising to 36.00 mg/L in March, and peaking at 57.75 mg/L in May. ANOVA (Analysis of Variance) indicates no significant differences in TDS levels among the study stations ($P > 0.05$).

4.1.6 Dissolved Oxygen (mg/L)

The spatial and temporal variations in dissolved oxygen are presented in Fig. 4.6. Spatially, station 4 had the highest mean concentration (6.67 mg/L), followed by station 2 (6.60 mg/L) and station 1 (6.53 mg/L), while station 3 recorded the lowest (6.33 mg/L). Temporally, the mean dissolved oxygen rose across the sampling months, from 6.33 mg/L in January to 6.60 mg/L in March, peaking at 6.67 mg/L in May. ANOVA showed no significant differences among the stations ($P > 0.05$).

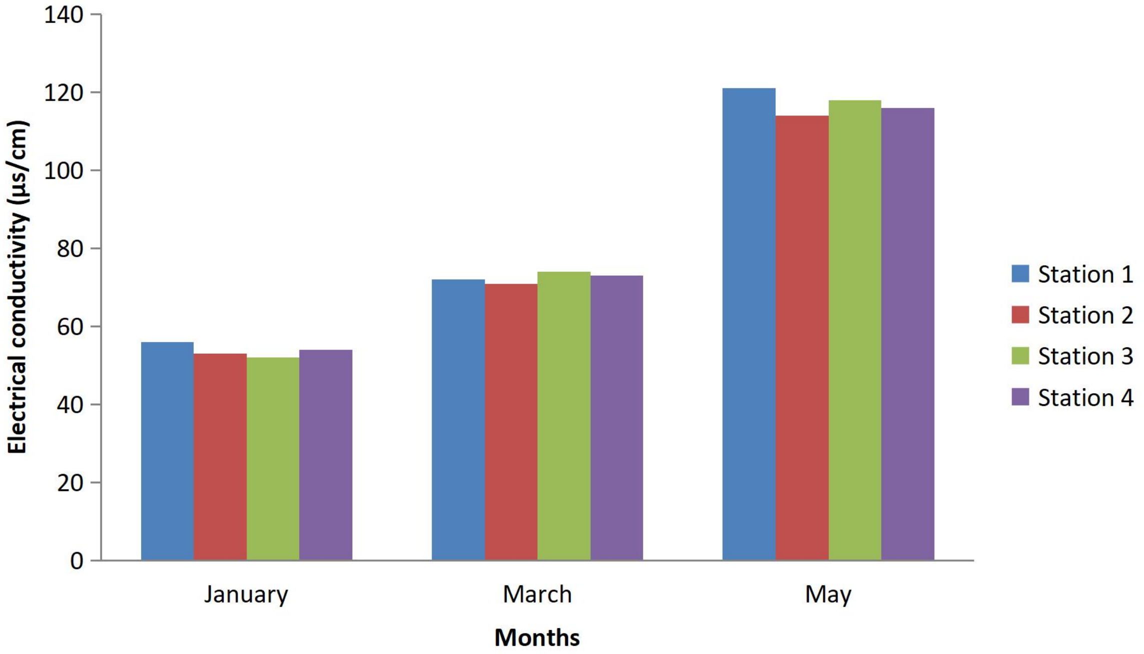

Figure 4.3: Spatial and temporal variations in Electrical Conductivity at the study stations of Ovia River, Benin City

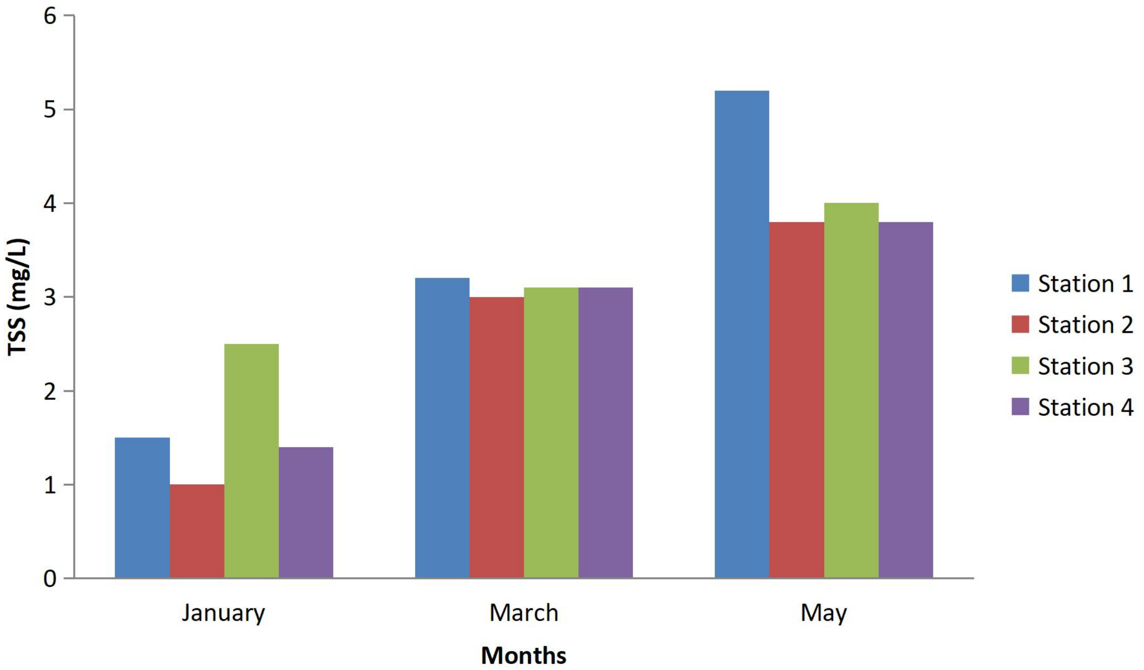

Figure 4.4: Spatial and temporal variations in Total Suspended Solids (mg/L) at the study stations of Ovia River, Benin City

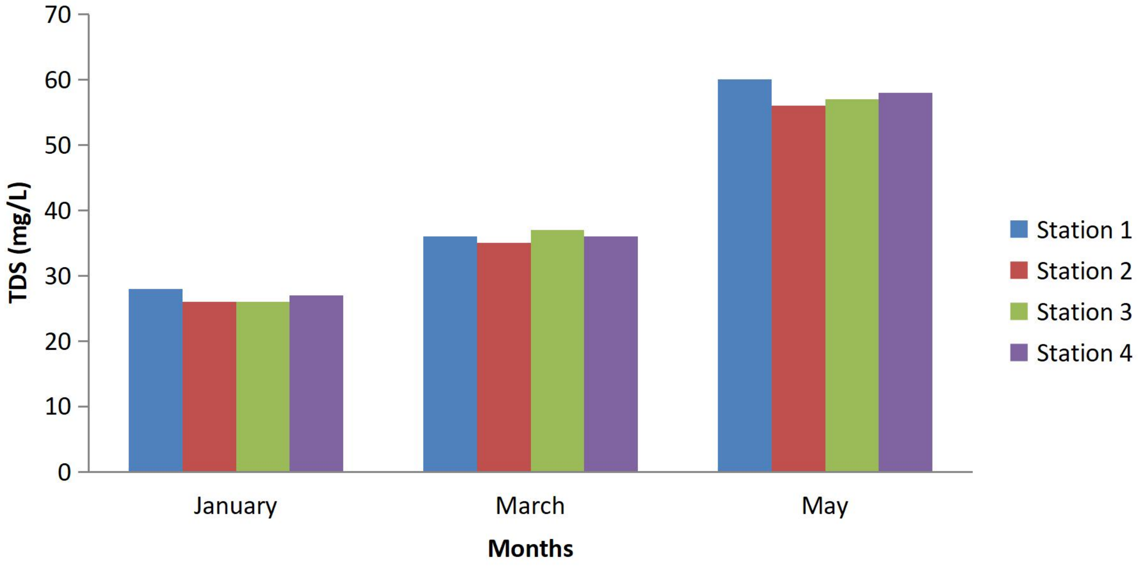

Figure 4.5: Spatial and temporal variations in Total dissolved Solids (mg/l) at the study stations of Ovia River, Benin City

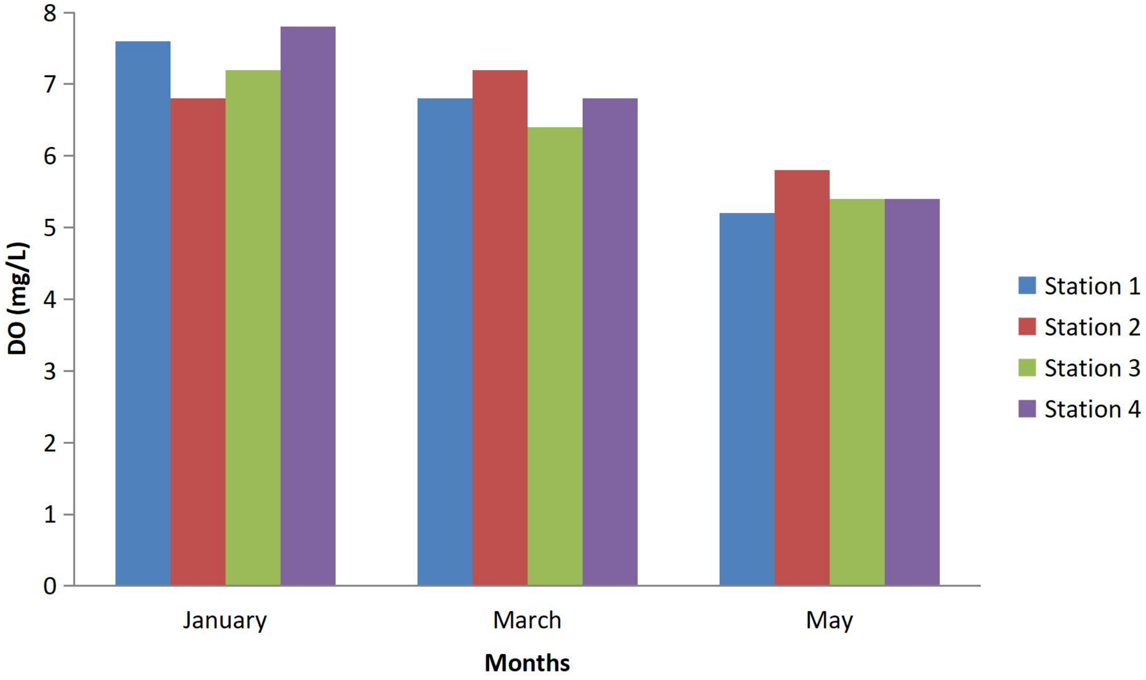

Figure 4.6: Spatial and temporal variations in Dissolved Oxygen (mg/L) at the study stations of Ovia River, Benin City

4.1.7 Biochemical Oxygen Demand (mg/L)

The spatial and temporary variations in biochemical oxygen demand are shown in Fig. 4.7. Station 1 recorded the highest mean value (3.60 mg/L), followed by station 4 (3.53 mg/L), station 2 (3.47 mg/L), and station 3 with the lowest (3.33 mg/L). Temporally, BOD increased across the months, from 3.23 mg/L in January to 3.53 mg/L in March, peaking at 3.73 mg/L in May. ANOVA revealed no significant differences among the stations ($P > 0.05$).

4.1.8 Chemical Oxygen Demand (mg/L)

The spatial and temporary variations in chemical oxygen demand measurements is shown in Fig. 4.8. Station 1 had the highest mean value (26.00 mg/L), followed by station 3 (23.33 mg/L), station 4 (22.67 mg/L), and station 2 with the lowest (20.33 mg/L). Temporally, COD rose steadily from 20.00 mg/L in January to 23.00 mg/L in March, reaching 26.33 mg/L in May. ANOVA showed no significant differences among the stations ($P > 0.05$).

4.1.9 Alkalinity (mg/L)

The spatial and temporal variations in alkalinity are illustrated in Fig. 4.9. Spatially, the highest mean alkalinity was recorded at station 1 and station 3, both with a value of 15.83 mg/L. This was followed by station 4, which recorded a mean alkalinity of 14.17 mg/L, the lowest mean was observed at station 2, with an average of 13.75 mg/L. Alkalinity values varied across the sampling months. The lowest mean was observed in January at 13.33 mg/L, which increased slightly to 14.58 mg/L in March, and peaked in May at 16.25 mg/L. ANOVA (Analysis of Variance) shows no significant differences in alkalinity levels among the study stations ($P > 0.05$).

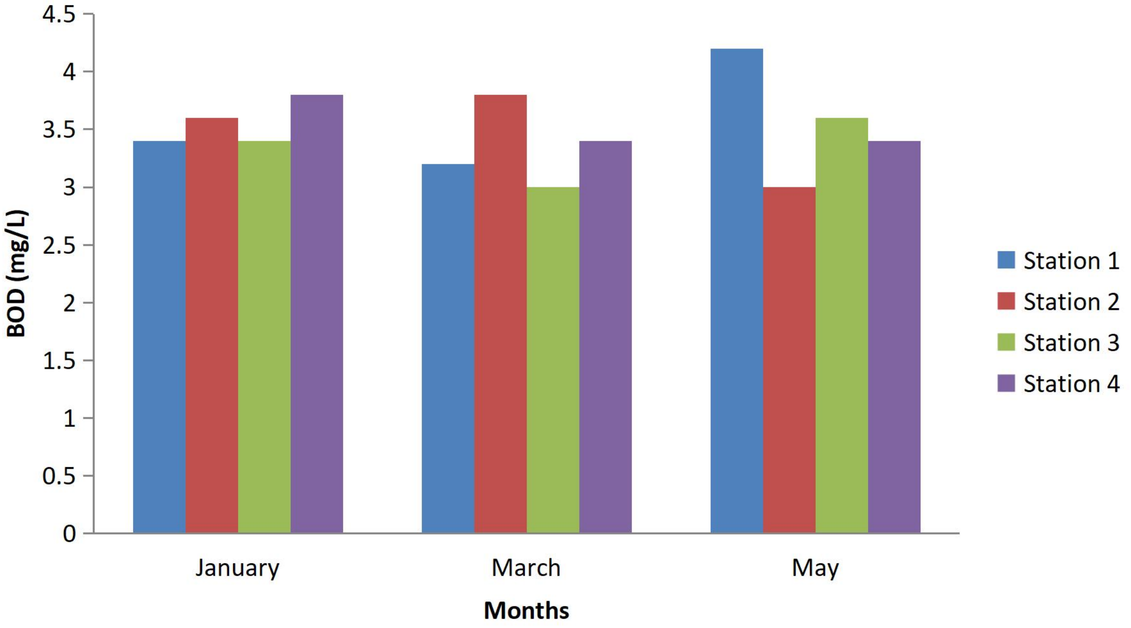

Figure 4.7: Spatial and temporal variations in Biochemical Oxygen Demand (mg/L) at the study stations of Ovia River, Benin City

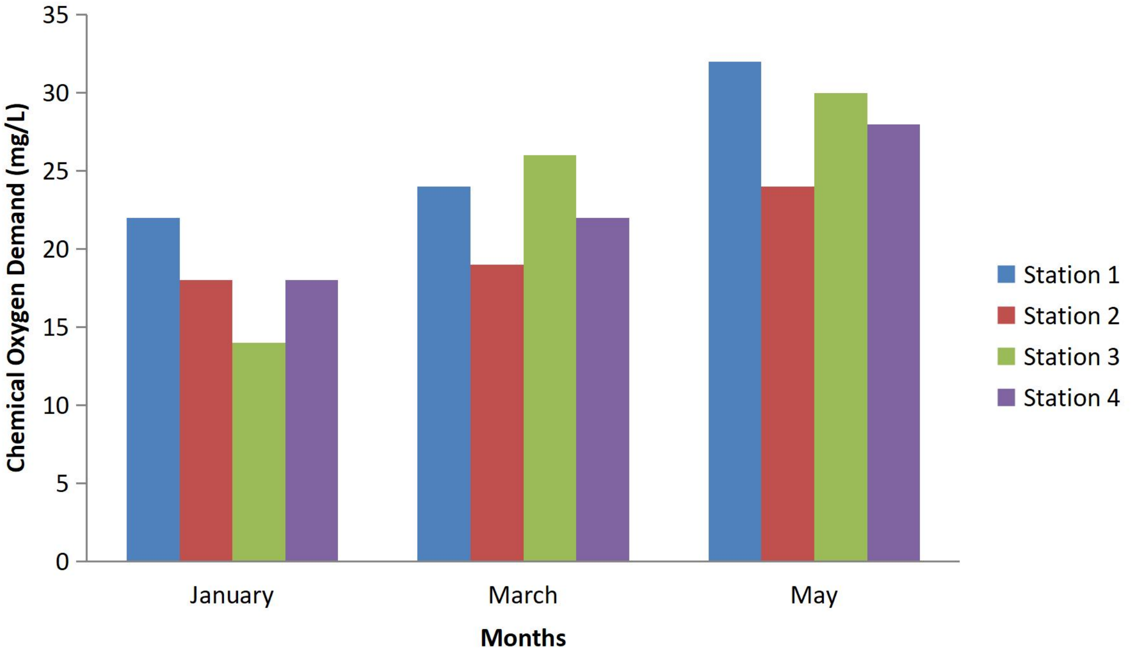

Figure 4.8: Spatial and temporal variations in Chemical Oxygen Demand (mg/L) at the study stations of Ovia River, Benin City

4.1.10 Turbidity (NTU)

The spatial and temporal variations in turbidity are presented in Fig. 4.10. Station 3 recorded the highest mean (8.49 NTU), followed by station 1 (8.32 NTU), station 4 (7.09 NTU), and station 2 with the lowest (6.06 NTU). Over time, turbidity rose from 5.70 NTU in January to 7.64 NTU in March, peaking at 9.06 NTU in May. ANOVA indicated no significant differences among stations ($P > 0.05$).

4.1.11 Sodium (Na^+)

The spatial and temporal variations in sodium concentration are illustrated in Fig. 4.11. Spatially, the highest mean concentration was observed at station 4 with 5.56 mg/L, followed closely by station 1 at 5.35 mg/L and station 3 at 5.30 mg/L. The lowest mean was recorded at station 2, with 5.07 mg/L. Temporally, sodium levels increased across the sampling periods. The mean was 4.68 mg/L in January, rising to 5.60 mg/L in March, and further to 5.95 mg/L in May. ANOVA (Analysis of Variance) indicates no significant differences in sodium levels among the study stations ($P > 0.05$)

4.1.12 Magnesium (Mg^{2+})

The spatial and temporary variations in magnesium measurements are shown in Fig. 4.12. The highest mean was recorded at station 4 with 2.92 mg/L, followed by station 1 at 2.85 mg/L. Station 3 had a mean of 2.66 mg/L, while station 2 exhibited the lowest mean of 2.59 mg/L. Temporally, the mean was 2.13 mg/L in January, increasing to 2.84 mg/L in March, and reaching 3.15 mg/L in May. ANOVA (Analysis of Variance) reveals no significant differences in magnesium levels across the study stations ($P > 0.05$).

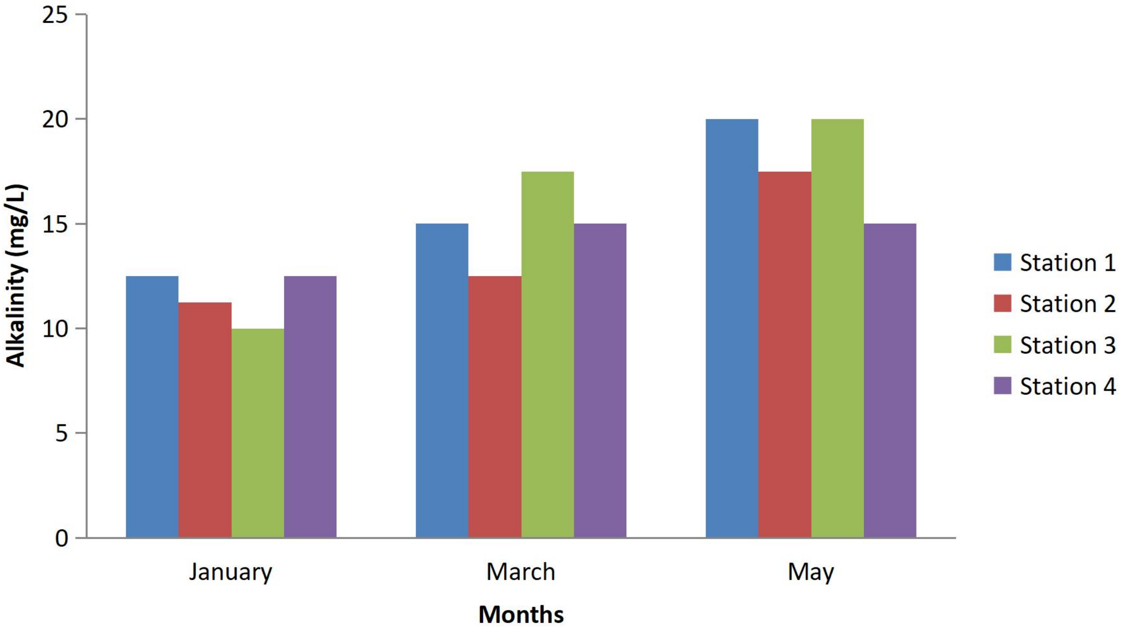

Figure 4.9: Spatial and temporal variations in Alkalinity (mg/L) at the study stations of Ovia River, Benin City

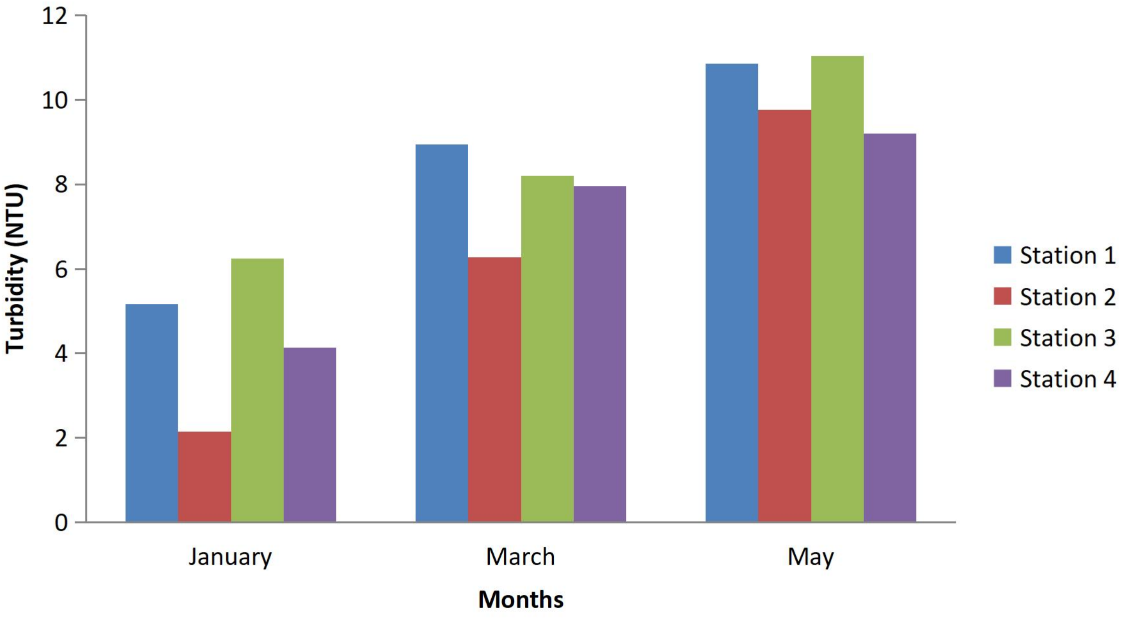

Figure 4.10: Spatial and temporal variations in Turbidity (NTU) at the study stations of Ovia River, Benin City

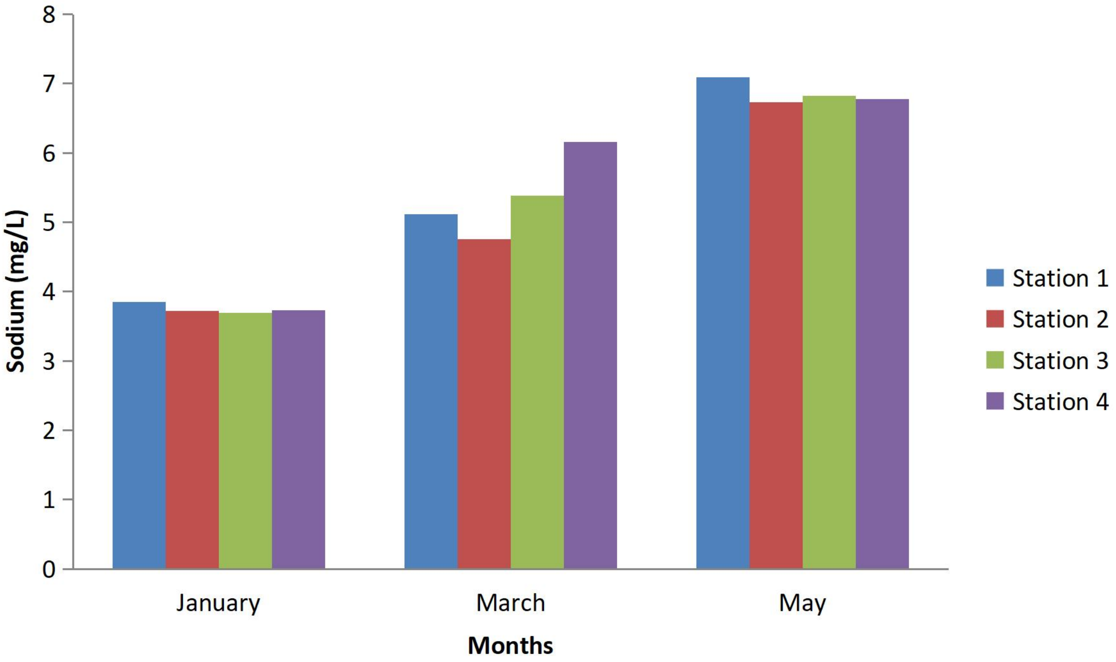

Figure 4.11: Spatial and Temporal Variations in Sodium at the study stations of Ovia River, Benin City

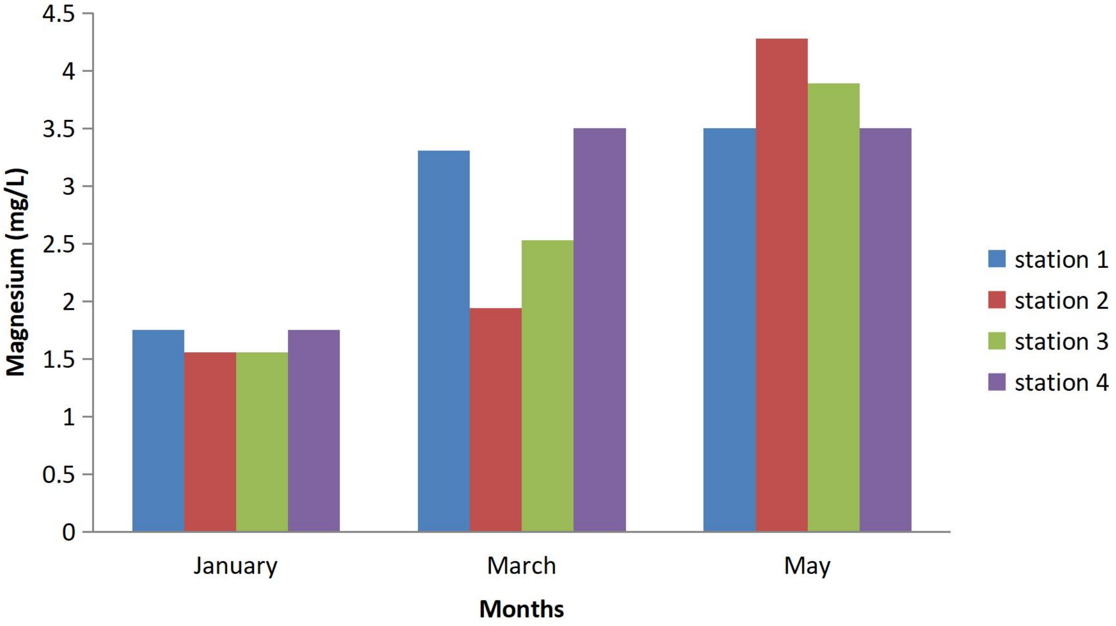

Figure 4.12: Spatial and temporal variations in Magnesium at the study stations of Ovia River, Benin City

4.1.13 Calcium (Ca^+)

The spatial and temporal variations in calcium concentration are presented in Fig. 4.13. Spatially, the highest mean concentration was recorded at both station 1 and station 2, each with 5.88 mg/L. This was followed by station 4, which had a slightly lower mean of 5.45 mg/L, and station 3, which recorded the lowest mean at 5.35 mg/L. Calcium levels increased progressively over time. The mean was 4.02 mg/L in January, rising to 5.90 mg/L in March, and peaking at 7.88 mg/L in May. ANOVA (Analysis of Variance) indicates no significant differences in calcium levels across the study stations ($P > 0.05$).

4.1.14 Potassium (K^+)

The spatial and temporary variations in potassium measurements are depicted in Fig. 4.14. Station 1 had the highest mean (3.72 mg/L), followed by station 3 (3.66 mg/L), station 4 (3.54 mg/L), and station 2 with the lowest (3.49 mg/L). Across months, values rose from 3.07 mg/L in January to 3.63 mg/L in March, peaking at 4.18 mg/L in May. ANOVA showed no significant differences among stations ($P > 0.05$).

4.1.15 Hardness (mg/L)

The spatial and temporary variations in hardness measurements are provided in Fig. 4.15. The highest mean concentration was recorded at station 1 with 26.42 mg/L, followed by station 4 with 25.62 mg/L. Station 2 had a slightly lower mean of 25.35 mg/L, while station 3 recorded the lowest mean at 24.29 mg/L. Temporally, hardness increased over time, with a mean of 18.38 mg/L in January, rising to 25.52 mg/L in March, and reaching a peak of 32.36 mg/L in May. The ANOVA indicates no significant differences in hardness levels across the study stations ($P > 0.05$).

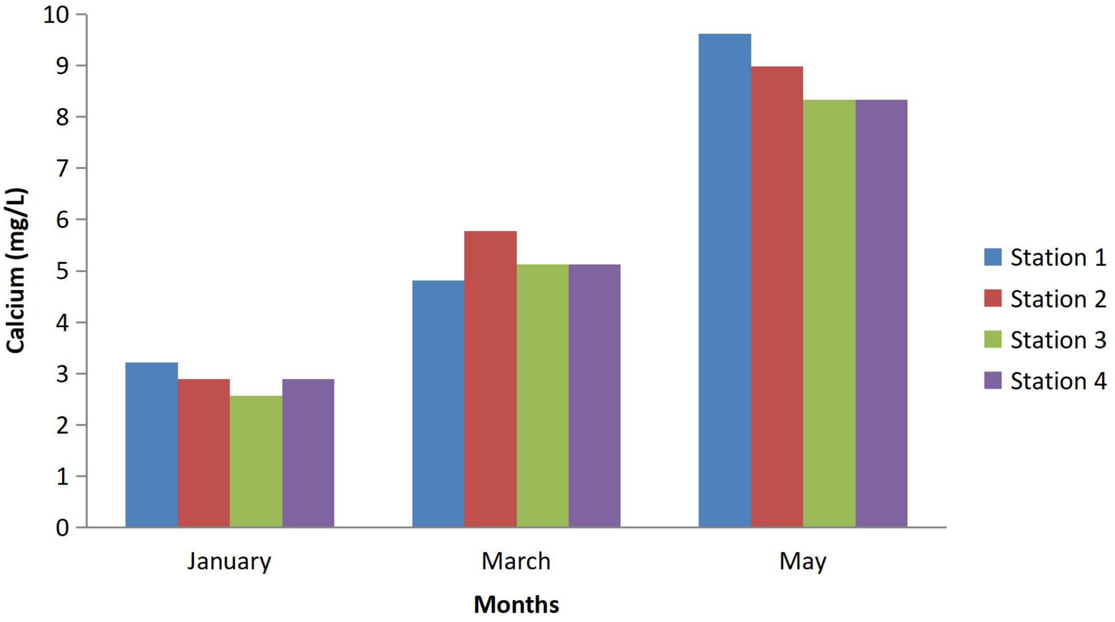

Figure 4.13: Spatial and temporal variations in Calcium at the study stations of Ovia River, Benin City

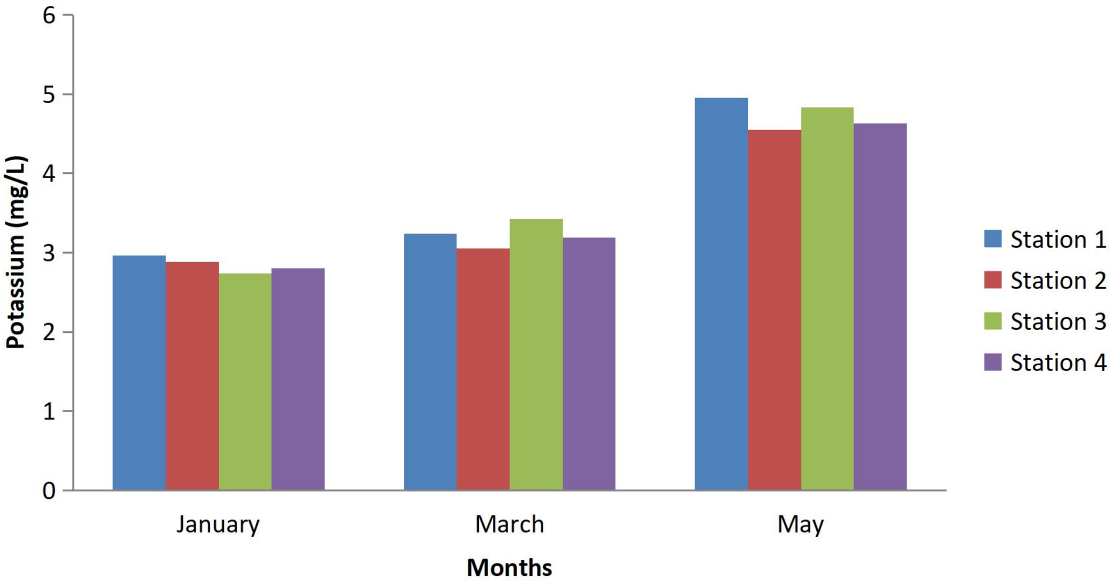

Figure 4.14: Spatial and temporal variations in Potassium at the study stations of Ovia River, Benin City

4.1.16 Chloride (Cl^-)

The spatial and temporary variations in chloride are represented in Fig. 4.16. Station 1 had the highest mean at 23.15 mg/L, followed by station 3 with 22.15 mg/L. Station 4 recorded a mean of 20.68 mg/L, while station 2 had the lowest mean at 20.09 mg/L. Temporally, chloride levels showed a steady increase from 16.94 mg/L in January to 22.02 mg/L in March, and further to 26.58 mg/L in May. ANOVA indicates no significant differences in chloride levels among the study stations ($P > 0.05$).

4.1.17 Sulphate (SO_4^{2-})

The spatial and temporal variations in sulphate concentrations are shown in Fig. 4.17. Station 4 recorded the highest mean value at 2.00 mg/L, followed by station 1 with a mean of 1.74 mg/L. Station 3 had a slightly lower concentration of 1.50 mg/L, while station 2 recorded the lowest mean at 1.39 mg/L. Temporally, sulphate concentrations was highest with an average of 2.17 mg/L in March, 1.54 mg/L in May, and lowest with 1.26 mg/L in January. ANOVA result indicates no significant differences in sulfate levels across the study stations ($P > 0.05$).

4.1.18 Phosphate (PO_4^{3-})

The spatial and temporary variations in phosphate are presented in Fig. 4.18. The highest mean concentration was recorded at station 4, with an average of 0.17 mg/L, station 1 and station 3 both exhibited an average phosphate concentration of 0.14 mg/L. Station 2 reported the lowest mean phosphate level at 0.13 mg/L. Phosphate concentrations was highest with an average of 0.18 mg/L in March, 0.16 mg/L in May, and lowest with 0.10 mg/L in January. ANOVA indicates no significant differences in phosphate levels across the study stations ($P > 0.05$).

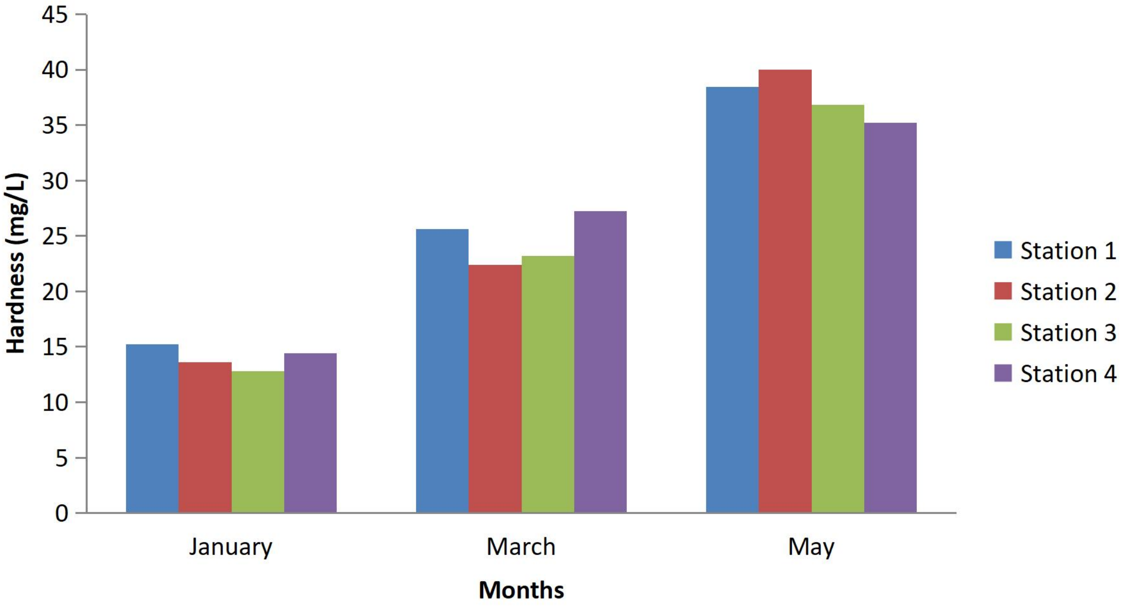

Figure 4.15: Spatial and temporal variations in Hardness at the study stations of Ovia River, Benin City

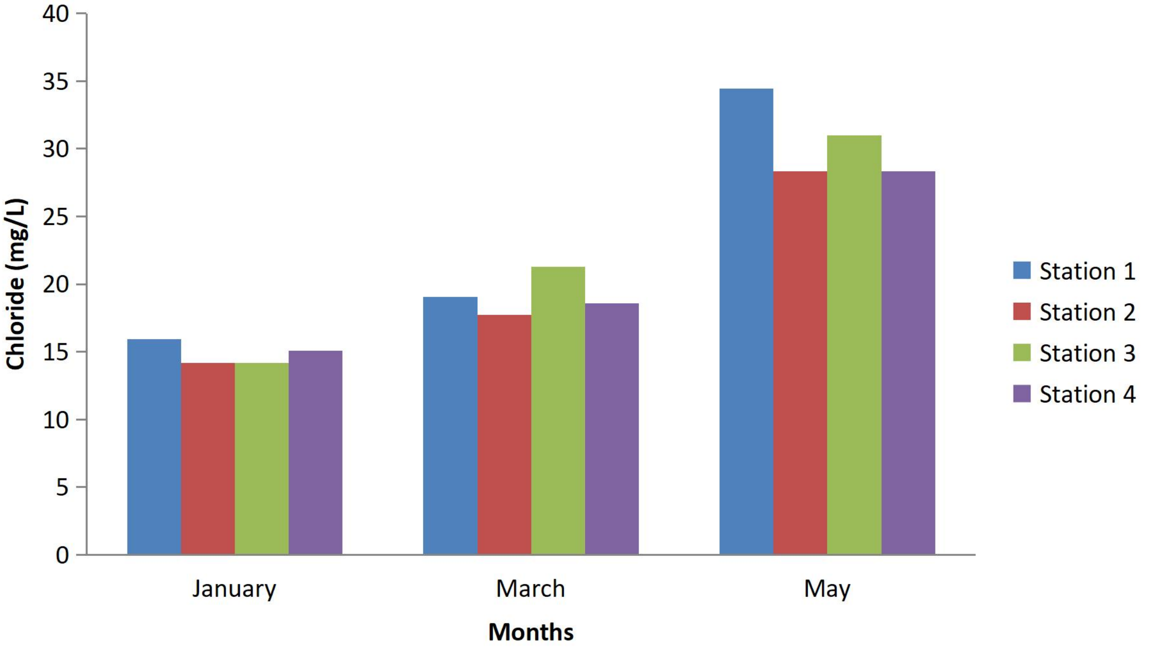

Figure 4.16: Spatial and temporal variations in Chloride at the study stations of Ovia River, Benin City

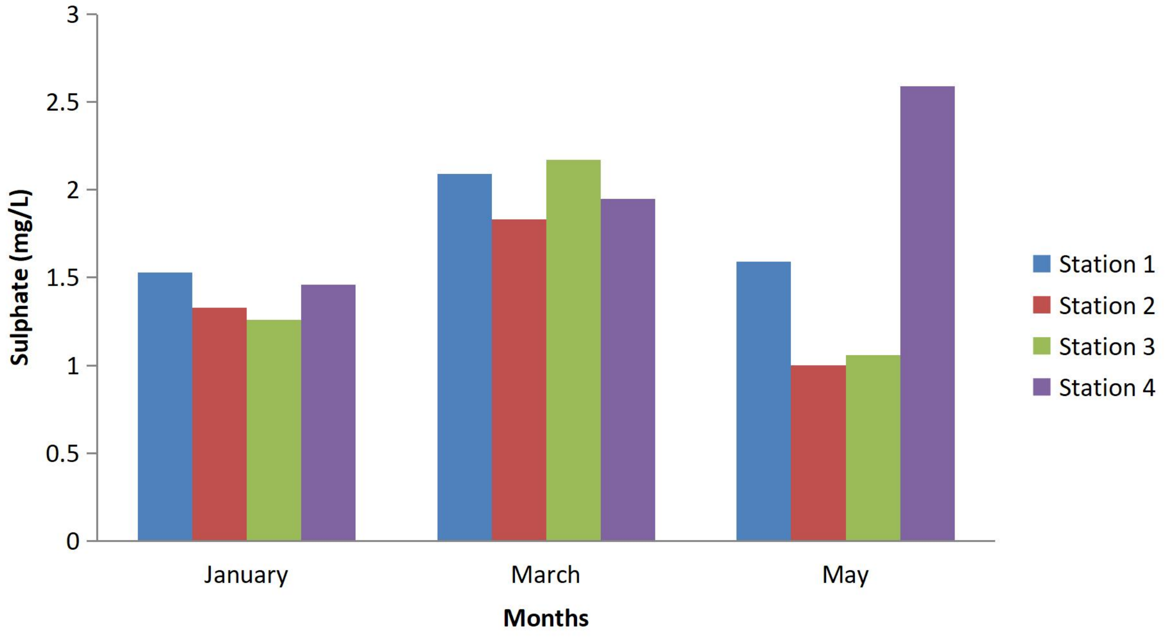

Figure 4.17: Spatial and Temporal Variations in Sulphate at the study stations of Ovia River, Benin City

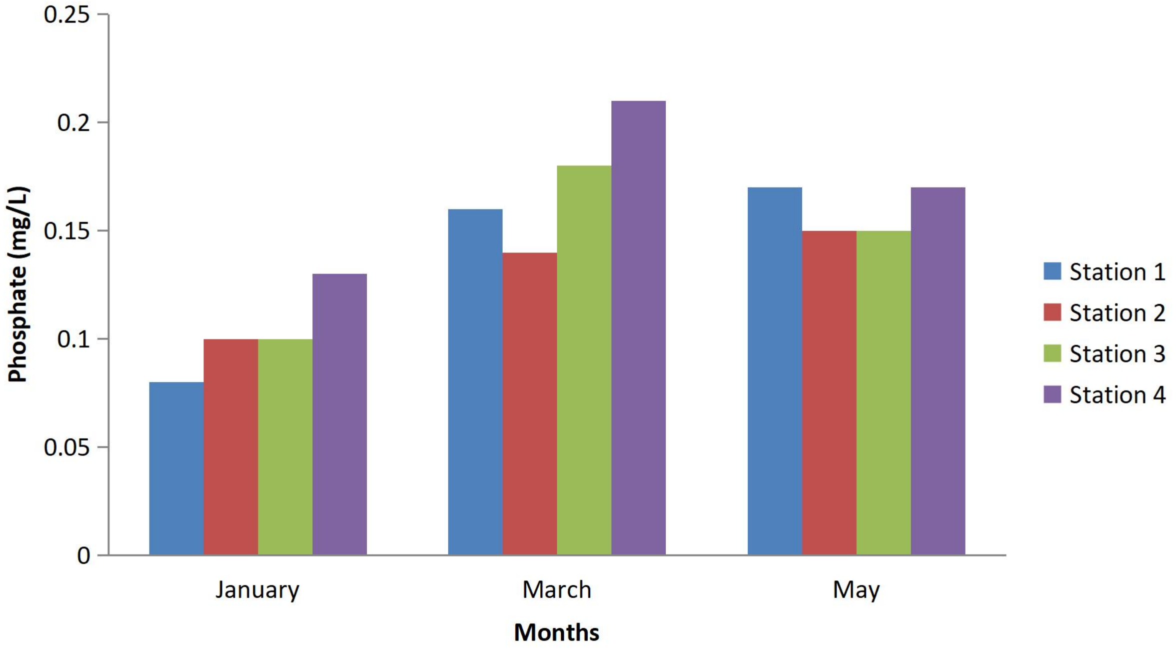

Figure 4.18: Spatial and temporal variations in Phosphate at the study stations of Ovia River, Benin City

4.1.19 Nitrate (NO_3^-)

The spatial and temporary variations in nitrate measurements are shown in Fig. 4.19. Station 4 had the highest mean (3.03 mg/L), followed by station 3 (2.68 mg/L), station 1 (2.67 mg/L), and station 2 with the lowest (2.53 mg/L). Across months, values were stable, with slightly higher means in May (2.68 mg/L) and January (2.67 mg/L), and a lower value in March (2.53 mg/L). ANOVA showed no significant differences among stations ($P > 0.05$).

4.1.20 Chromium (mg/L)

The spatial and temporal variations in chromium concentrations is presented in Fig. 4.20. The highest mean chromium levels were observed at stations 1 and 3, both recording a mean concentration of 0.03 mg/L. In contrast, stations 2 and 4 showed lower mean concentrations of 0.01 mg/L. Temporally, chromium concentrations were lowest in January (0.013 mg/L), increased slightly in March (0.018 mg/L), and peaked in May (0.025 mg/L)). The ANOVA indicates no significant differences in chromium levels across the study stations ($P > 0.05$).

4.1.21 Copper (mg/L)

The spatial and temporal variations in copper concentrations are illustrated in Fig. 4.21. The highest mean copper concentration was observed at station 3, with 0.04 mg/L. Station 2 followed with a mean of 0.02 mg/L, while stations 1 and 4 recorded the lowest average concentrations at 0.01 mg/L. Temporally, copper levels were lowest in January (0.01 mg/L) but increased notably in March and remained stable through May (both at 0.025 mg/L). ANOVA indicates significant differences in copper levels across the study stations ($P < 0.05$).

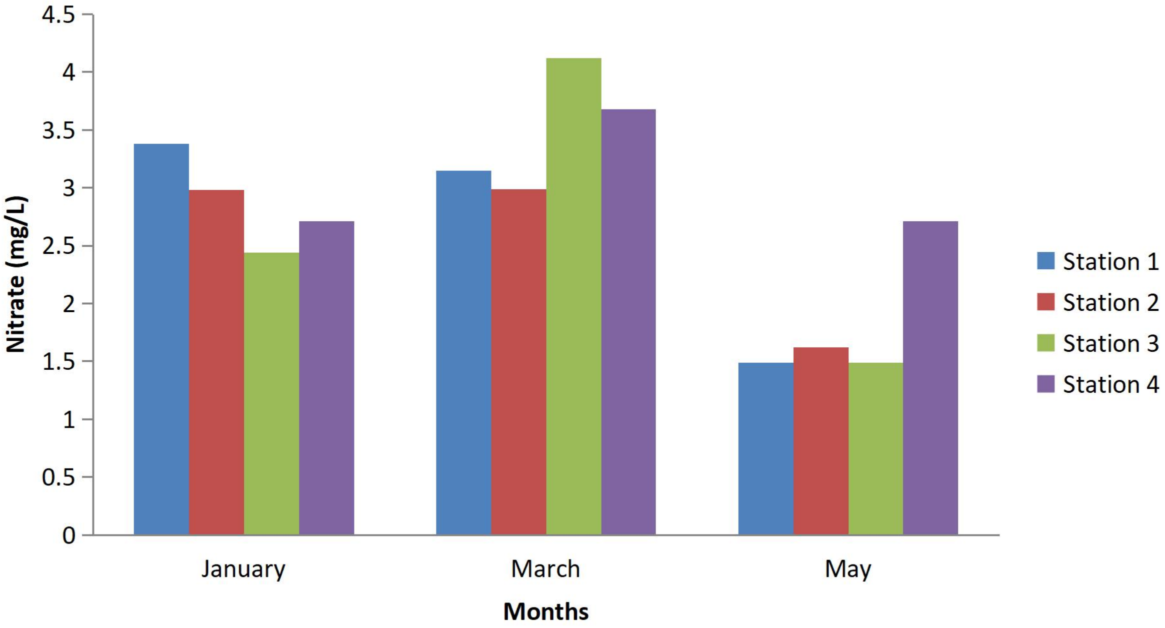

Figure 4.19: Spatial and temporal variations in Nitrate at the study stations of Ovia River, Benin City

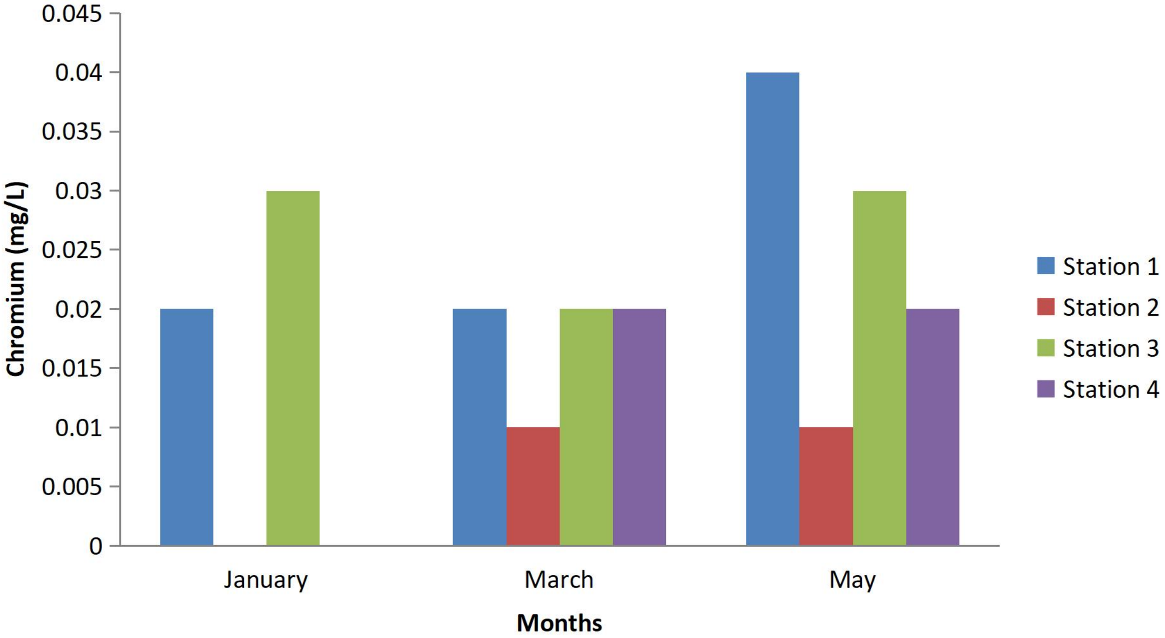

Figure 4.20: Spatial and temporal variations in Chromium at the study stations of Ovia River, Benin City

4.1.22 Iron (mg/L)

The spatial and temporary variations in iron measurements are shown in Fig. 4.22. Station 3 recorded the highest mean iron concentration at 0.26 mg/L, followed by station 4 with 0.20 mg/L. Station 2 reported a moderate mean concentration of 0.15 mg/L, while station 1 had the lowest at 0.14 mg/L. Temporally, iron concentrations were highest in March (0.235 mg/L), slightly lower in January (0.20 mg/L), and declined markedly in May (0.125 mg/L). ANOVA indicates no significant differences in iron levels among the stations ($P > 0.05$).

4.1.23 Manganese (mg/L)

The spatial and temporary variations in manganese measurements are shown in Fig. 4.23. The highest mean concentration was found at station 3, averaging 0.04 mg/L. Stations 1, 2, and 4 all recorded lower mean concentrations of 0.02 mg/L. Temporally, manganese levels peaked in March (0.03 mg/L), followed by January (0.025 mg/L), and declined in May (0.018 mg/L). The statistical analysis indicates significant differences in manganese levels across the study stations ($P < 0.05$).

4.1.24 Nickel (mg/L)

The spatial and temporary variations in nickel measurements are represented in Fig. 4.24. Station 3 recorded the highest mean concentration at 0.02 mg/L, consistent across all sampling periods while stations 1, 2, and 4 each had lower mean concentrations of 0.01 mg/L. Temporally, nickel concentrations were lowest in January (0.0075 mg/L), but increased in March and remained steady through May (both at 0.015 mg/L). ANOVA shows no significant differences in nickel levels across the stations ($P > 0.05$).

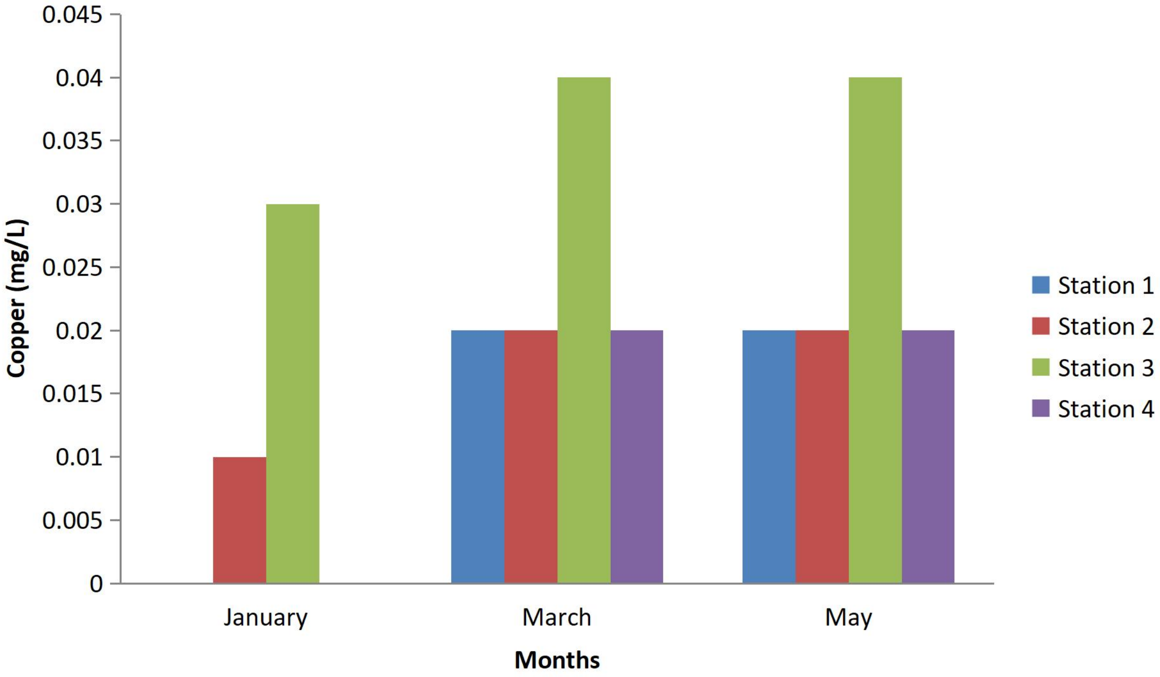

Figure 4.21: Spatial and temporal variations in Copper at the study stations of Ovia River, Benin City

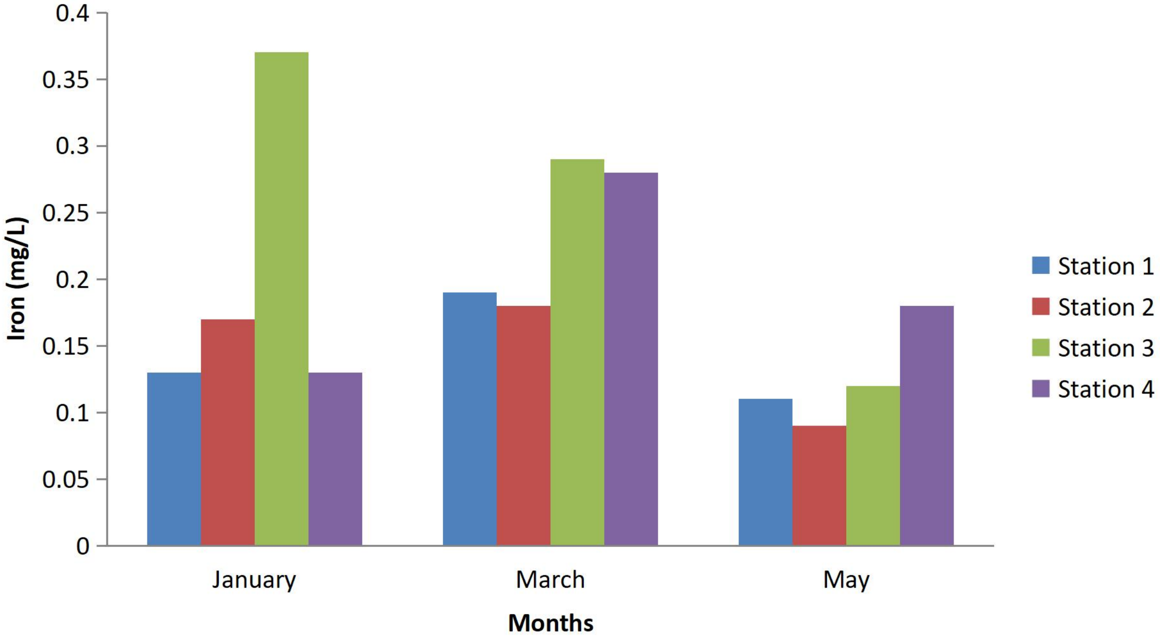

Figure 4.22: Spatial and temporal variations in Iron at the study stations of Ovia River, Benin City

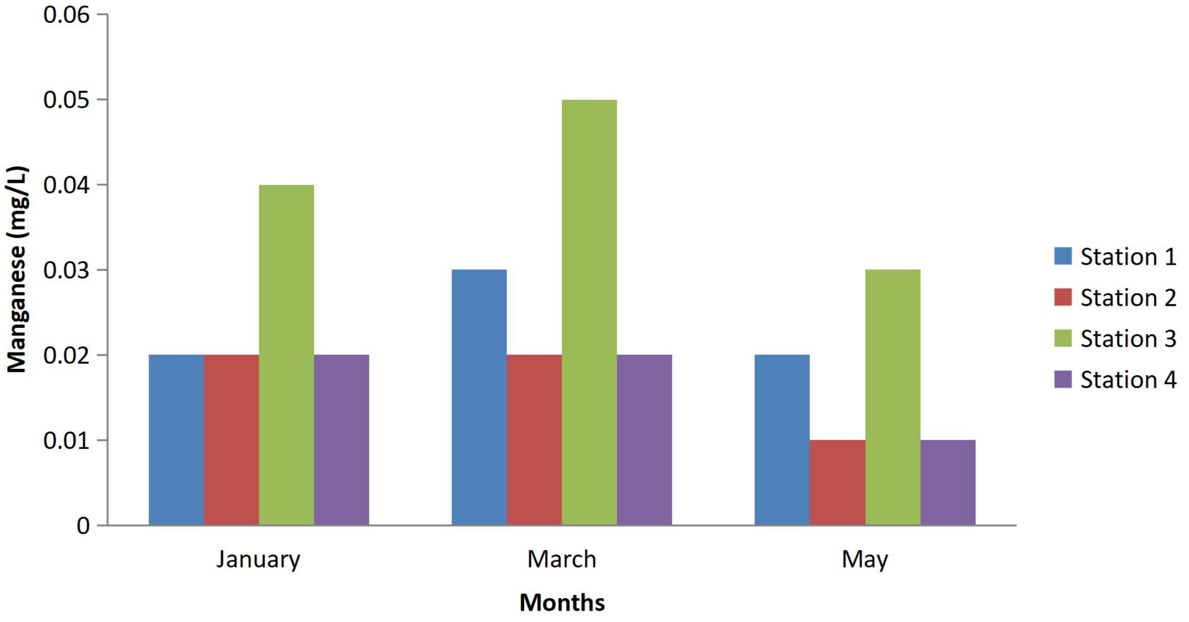

Figure 4.23: Spatial and temporal variations in Manganese at the study stations of Ovia River, Benin City

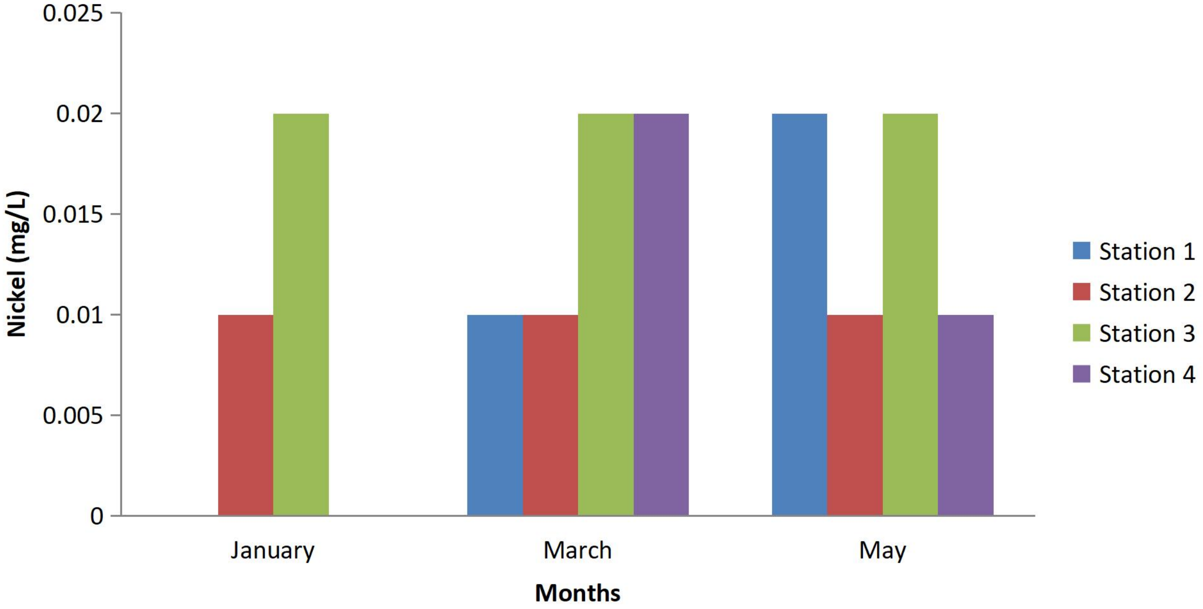

Figure 4.24: Spatial and temporal variations in Nickel at the study stations of Ovia River, Benin City

4.1.25 Lead (mg/L)

The spatial and temporary variations in lead measurements is shown in Fig. 4.25. The highest mean concentration was recorded at station 1 and station 3, both with a mean of 0.01 mg/L, and ranges extending from 0.00 mg/L to 0.02 mg/L for station 3. In contrast, stations 2 and 4 reported no detectable levels of lead, with mean concentration of 0.00 mg/L. Temporally, lead levels were slightly higher in May (0.0075 mg/L) compared to March and January (both at 0.005 mg/L). ANOVA indicates significant differences in lead levels across the study stations ($P < 0.05$).

4.1.26 Zinc (mg/L)

The spatial and temporary variations in zinc measurements are displayed in Fig. 4.26. The highest mean concentration was recorded at station 3, with a mean of 0.12 mg/L and a range from 0.09 mg/L to 0.12 mg/L. Station 1 followed with a mean of 0.08 mg/L, and values ranging from 0.05 mg/L to 0.10 mg/L. Station 2 reported a slightly lower mean concentration of 0.07 mg/L, with values spanning from 0.06 mg/L to 0.08 mg/L. Lastly, station 4 had the lowest mean level at 0.06 mg/L, with a range between 0.04 mg/L and 0.08 mg/L. Temporally, zinc levels peaked in March (0.09 mg/L), declined slightly in May (0.0775 mg/L), and were lowest in January (0.0675 mg/L). ANOVA indicates no significant differences in zinc levels across the study stations ($P > 0.05$).

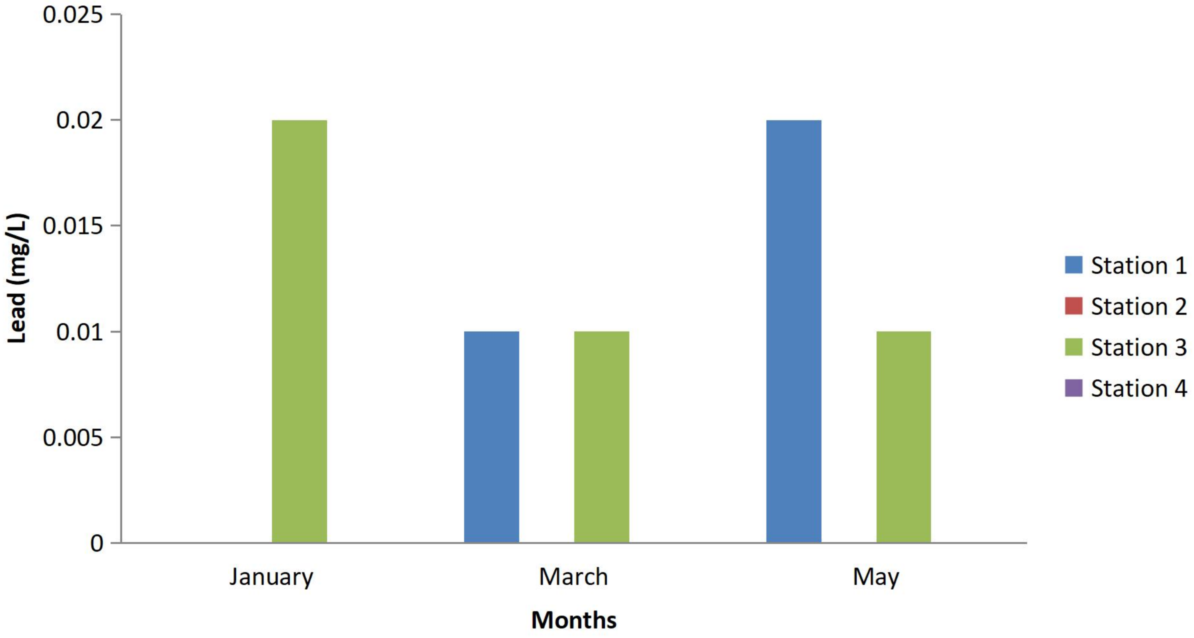

Figure 4.25: Spatial and temporal variations in Lead at the study stations of Ovia River, Benin City

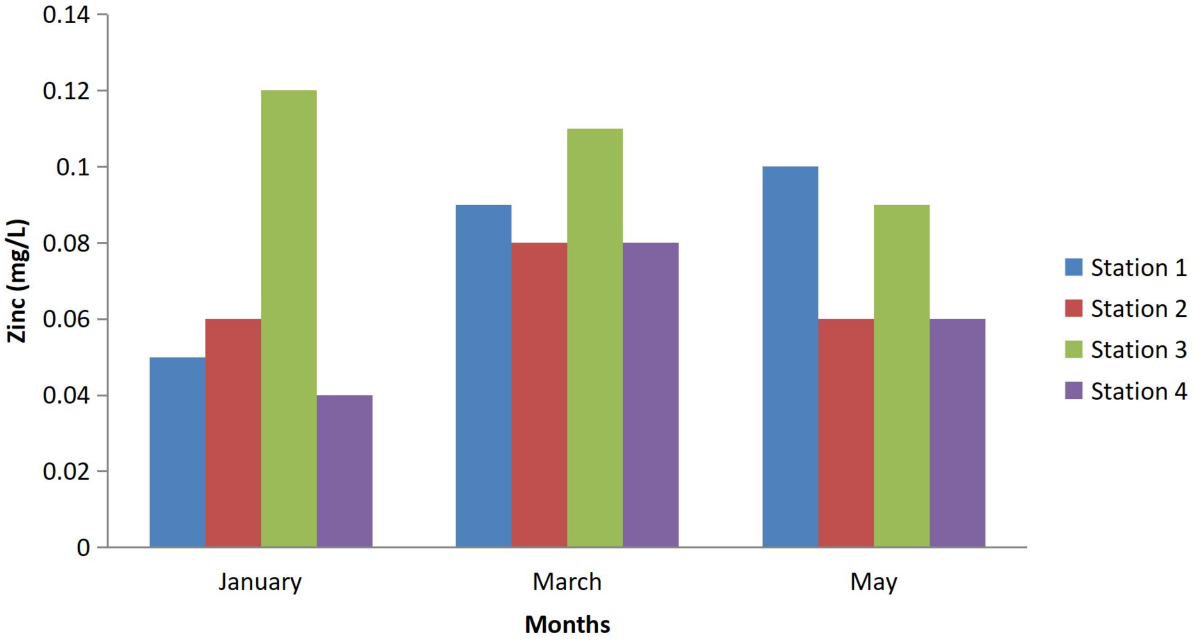

Figure 4.26: Spatial and temporal variations in Zinc at the study stations of Ovia River, Benin City

4.2 Microplastics in Surface Water

4.2.1 Particle Size Distribution (PSD) Analysis

The result of the particle size distribution (PSD) of microplastics in the surface water across four stations are presented in Table 4.2 and fig 4.27- 4.38. In January, Stations 1 and 2 recorded higher mean particle sizes (1.08 ± 0.80 and 1.73 ± 1.01, respectively), Stations 3 and 4 had comparatively lower mean values (1.01 ± 0.54 and 1.08 ± 0.57). In March, mean PSD values increased at Stations 1 and 2, with Station 2 again showing the highest mean (1.83 ± 2.61). Station 3 recorded the lowest mean PSD (0.52 ± 0.54). Station 4 remained moderate (0.83 ± 0.83). By May, a general decline in mean PSD was observed across all stations. Station 2 showed the most significant reduction (0.48 ± 0.55), while Station 4 recorded the highest mean (0.81 ± 0.80).

Table 4.2: Summary of PSD (µm) in the Surface Water of Ovia River, Benin City

Station	**January** **(Mean ± SD)**	**March** **(Mean ± SD)**	**May** **(Mean ± SD)**
Station 1	1.08 ± 0.80	1.78 ± 1.04	0.74 ± 1.53
Station 2	1.73 ± 1.09	1.83 ± 2.61	0.46 ± 0.55
Station 3	1.01 ± 0.54	0.52 ± 0.54	0.71 ± 1.14
Station 4	1.08 ± 0.56	0.83 ± 0.83	0.81 ± 0.80

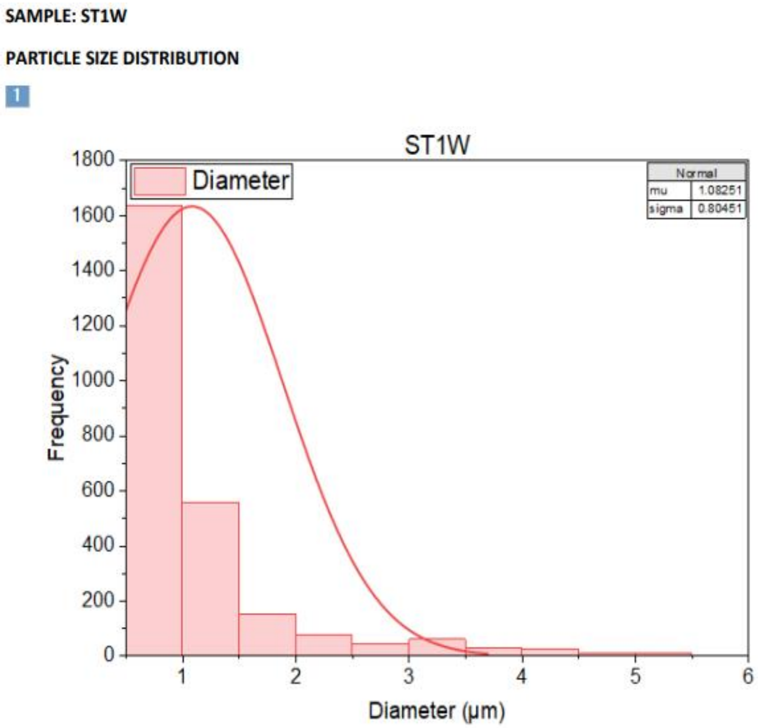

Figure 4.27: Particle Size Distribution of Microplastics at Station 1 (January).

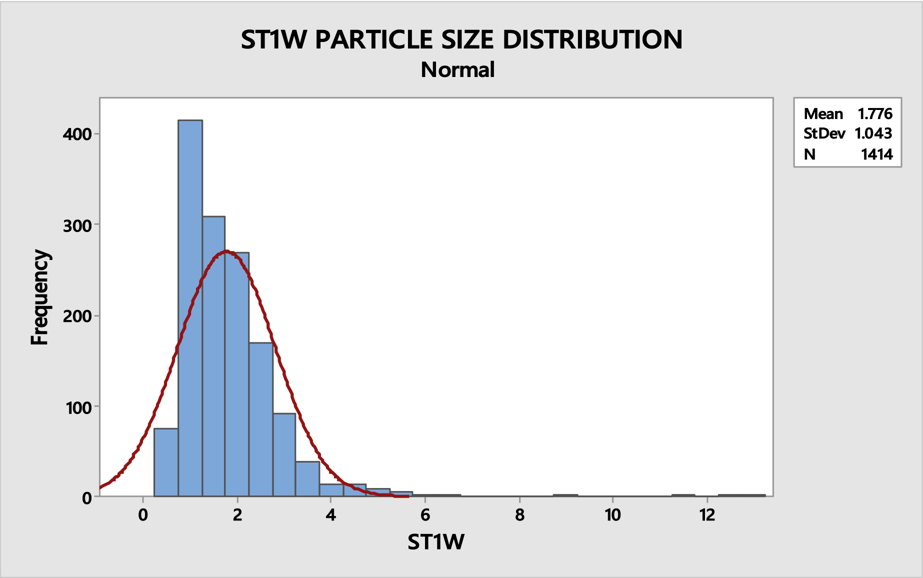

Figure 4.28: Particle Size Distribution of Microplastics at Station 1 (March).

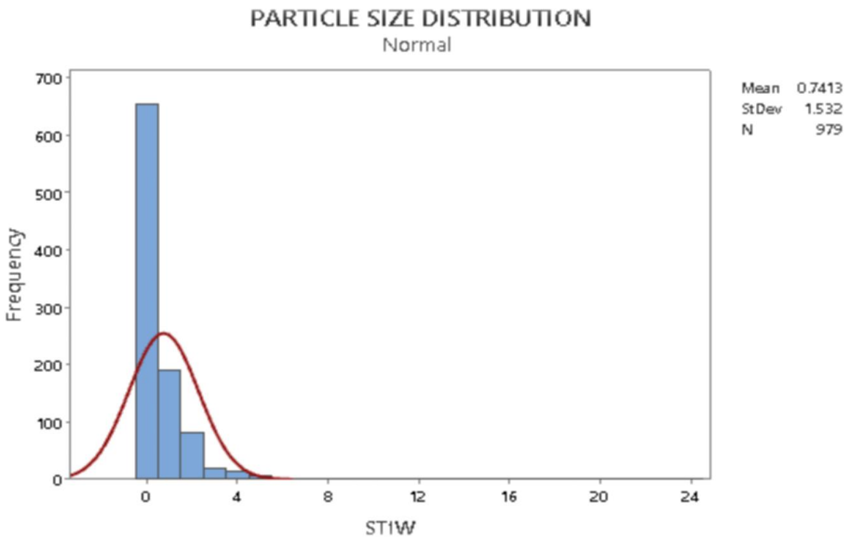

Figure 4.29: Particle Size Distribution of Microplastics at Station 1 (May).

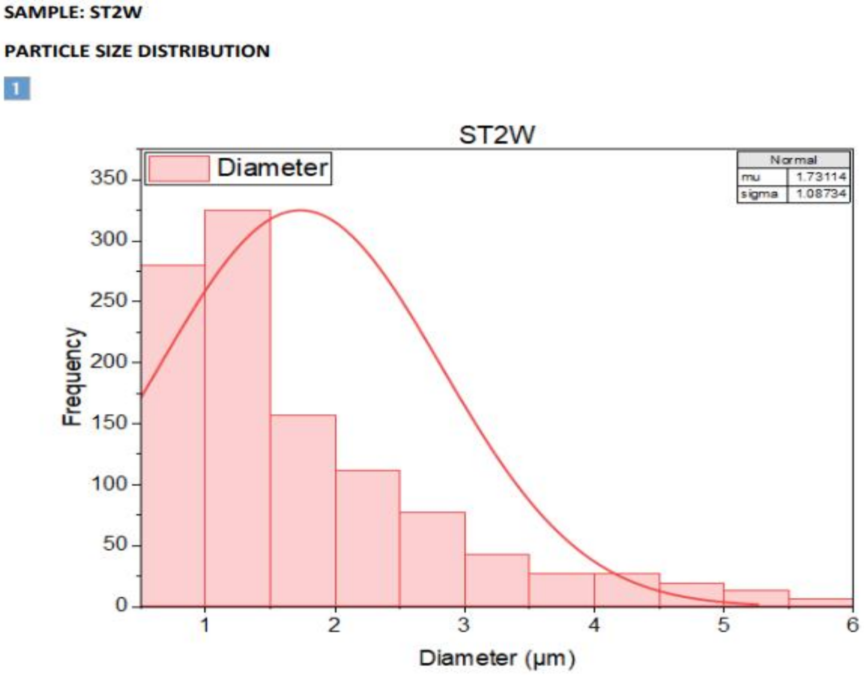

Figure 4.30: Particle Size Distribution of Microplastics at Station 2 (January).

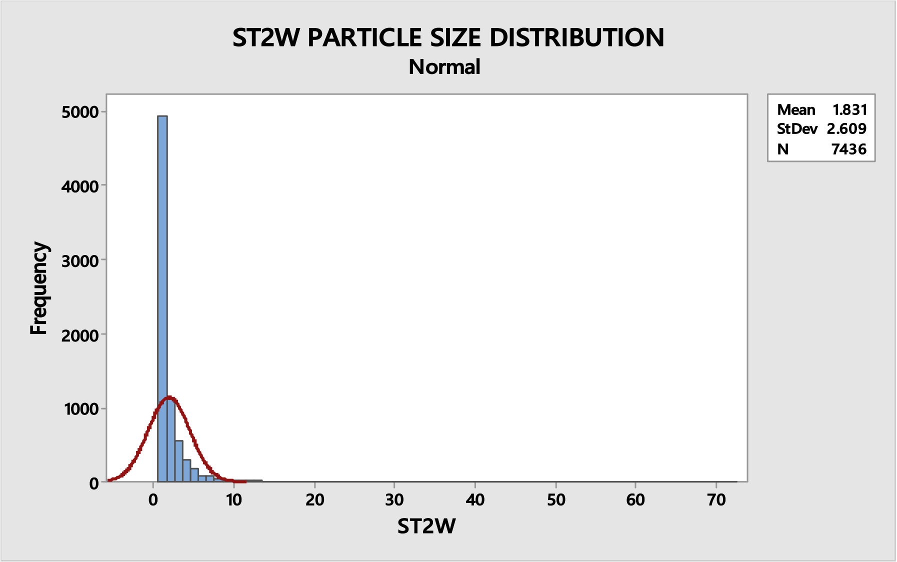

Figure 4.31: Particle Size Distribution of Microplastics at Station 2 (March).

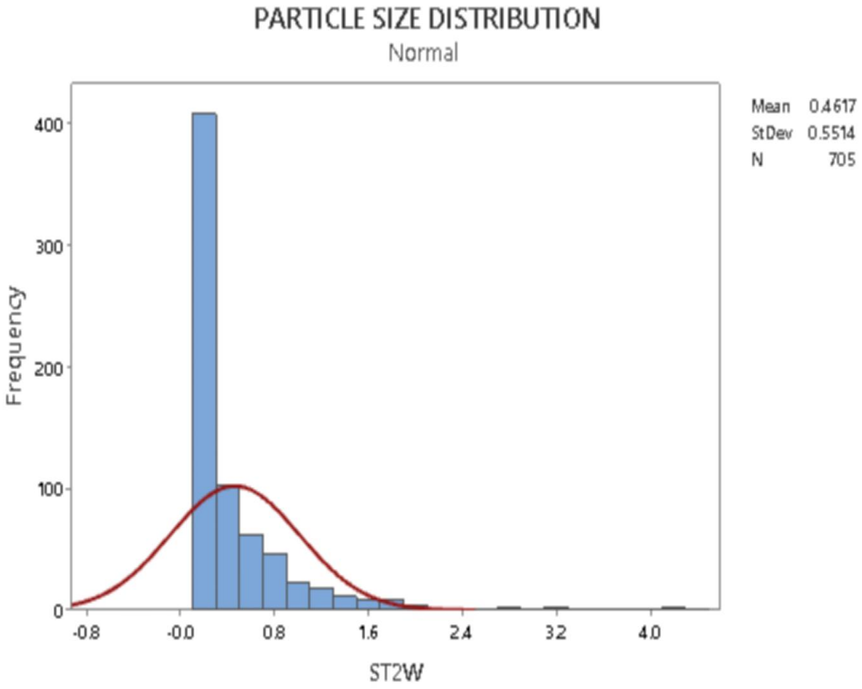

Figure 4.32: Particle Size Distribution of Microplastics at Station 2 (May).

SAMPLE: ST3W

PARTICLE SIZE DISTRIBUTION

1

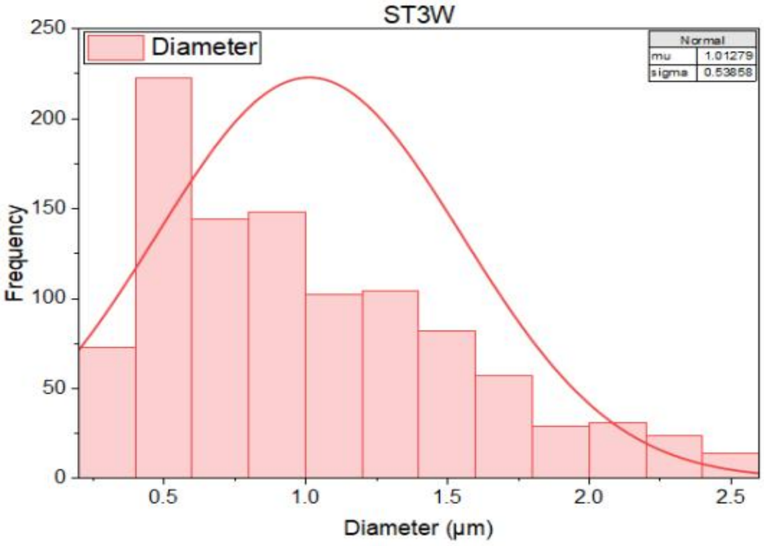

Figure 4.33: Particle Size Distribution of Microplastics at Station 3 (January).

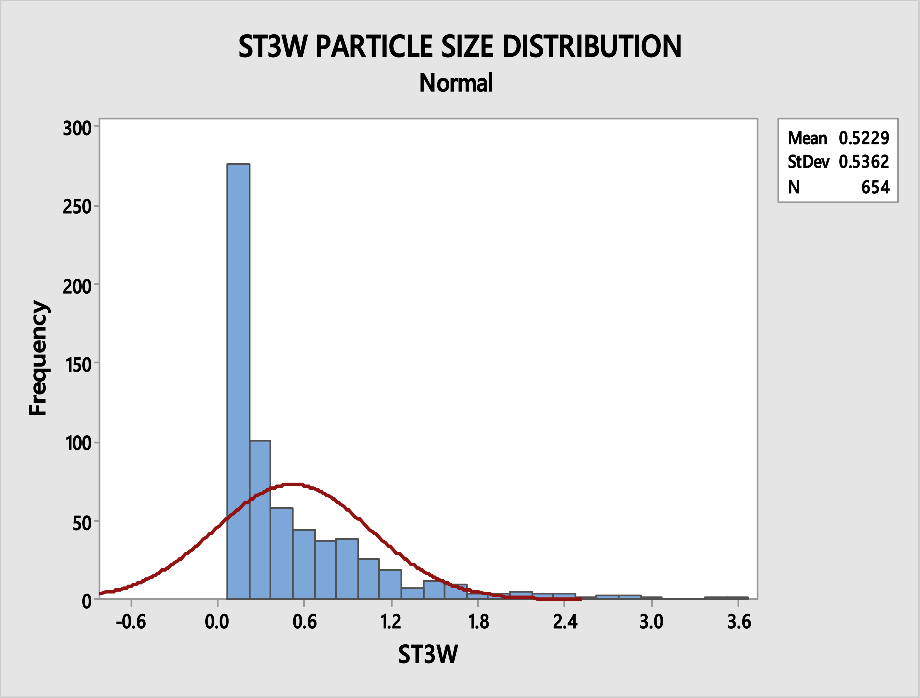

Figure 4.34: Particle Size Distribution of Microplastics at Station 3 (March).

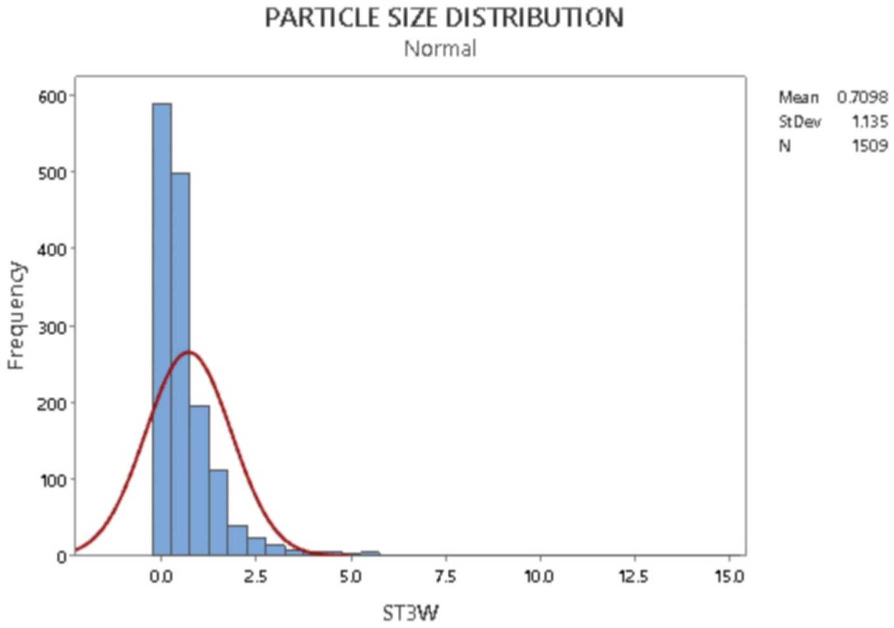

Figure 4.35: Particle Size Distribution of Microplastics at Station 3 (May).

SAMPLE: ST4W

PARTICLE SIZE DISTRIBUTION

1

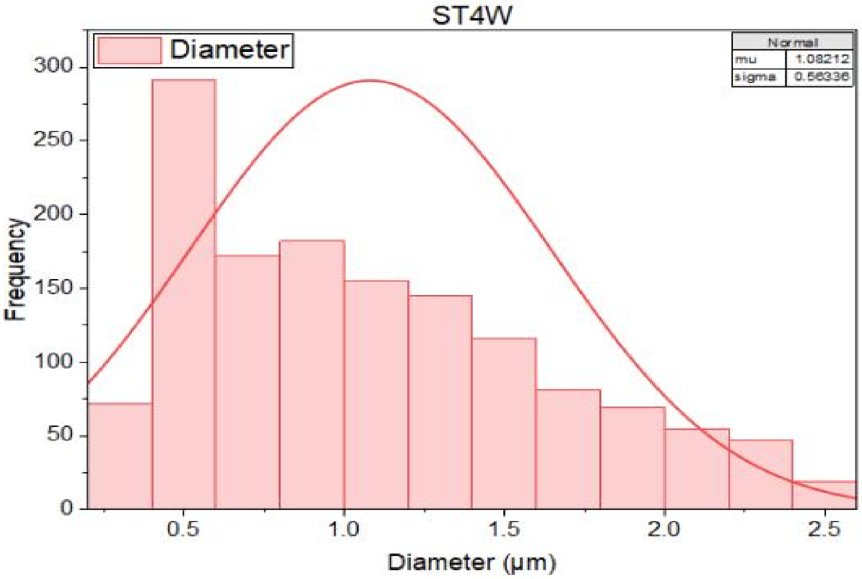

Figure 4.36: Particle Size Distribution of Microplastics at Station 4 (January)

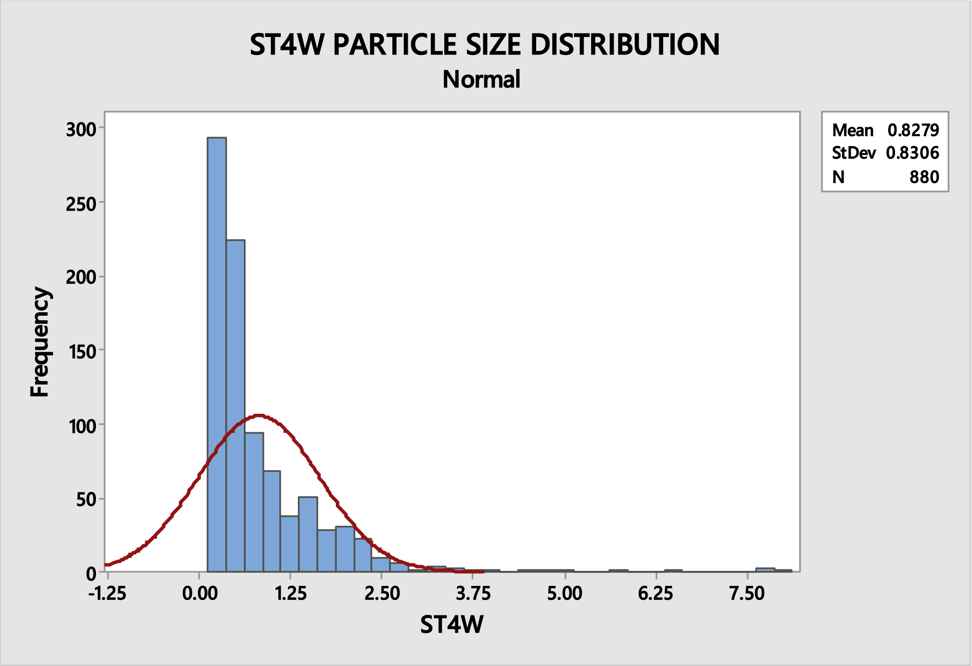

Figure 4.37: Particle Size Distribution of Microplastics at Station 4 (March)

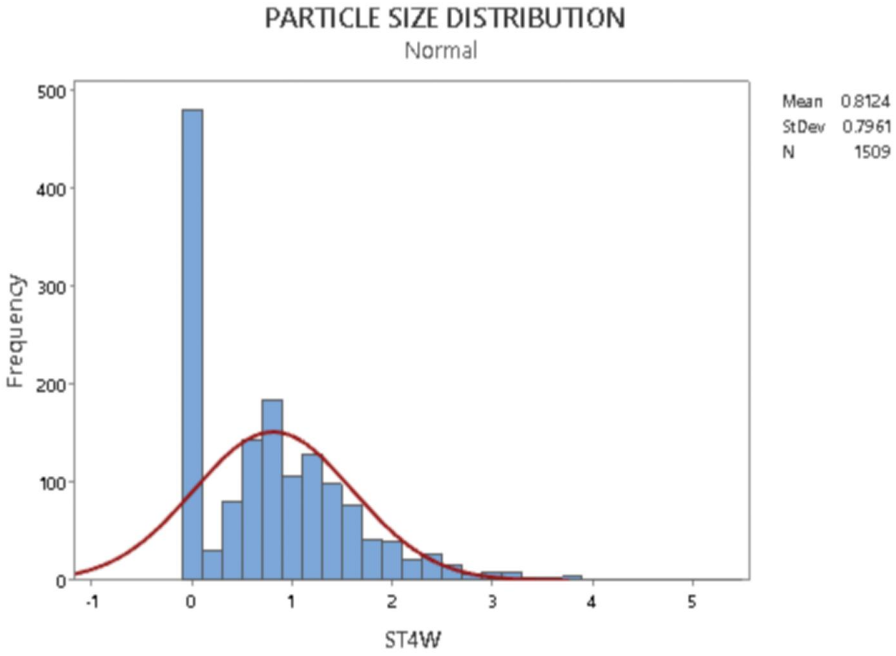

Figure 4.38: Particle Size Distribution of Microplastics at Station 4 (May).

4.2.2 Polymer Analysis of Microplastics in the Surface Water Samples

The FTIR spectra for microplastics particles extracted from surface water samples are showed in Fig. 4.39–4.50, while the FTIR results are summarized in the Table 4.3, showing the observed absorption peaks (in cm^{-1}), the corresponding polymer types, their characteristic peaks, and the functional group assignments. A total of 10 distinct polymers were identified across the study period, namely: polyvinyl alcohol (PVA), polyamide (nylon), polypropylene (PP), low-density polyethylene (LDPE), high-density polyethylene (HDPE), polyethylene terephthalate (PET), polyurethane, polyacetylene, polyethylene glycol (PEG), and cellophane. In terms of spatial distribution, four polymer types were detected at station 1, six at station 2, seven at station 3, and four at station 4. Notably, O–H stretching vibrations, indicative of alcohol or hydroxyl-containing groups (commonly found in PVA, PEG, and cellophane), and C=O stretching, characteristic of carbonyl groups in polyamides, polyurethanes, and polyesters (such as PET), were frequently observed. Other characteristic peaks included CH_2 and CH_3 stretching vibrations, associated with polyolefins like PP and LDPE, and C≡C stretching, attributed to polyacetylene.

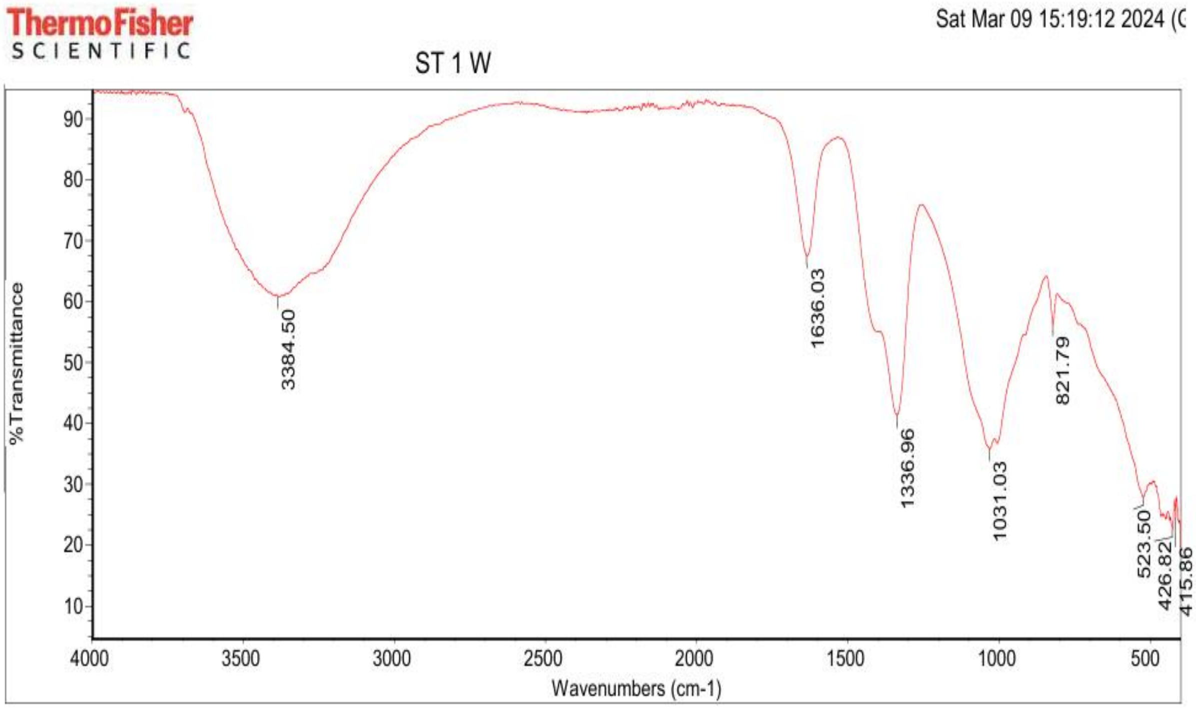

Fig 4.39: FTIR Spectra for Surface Water Sample in Station 1 (January)

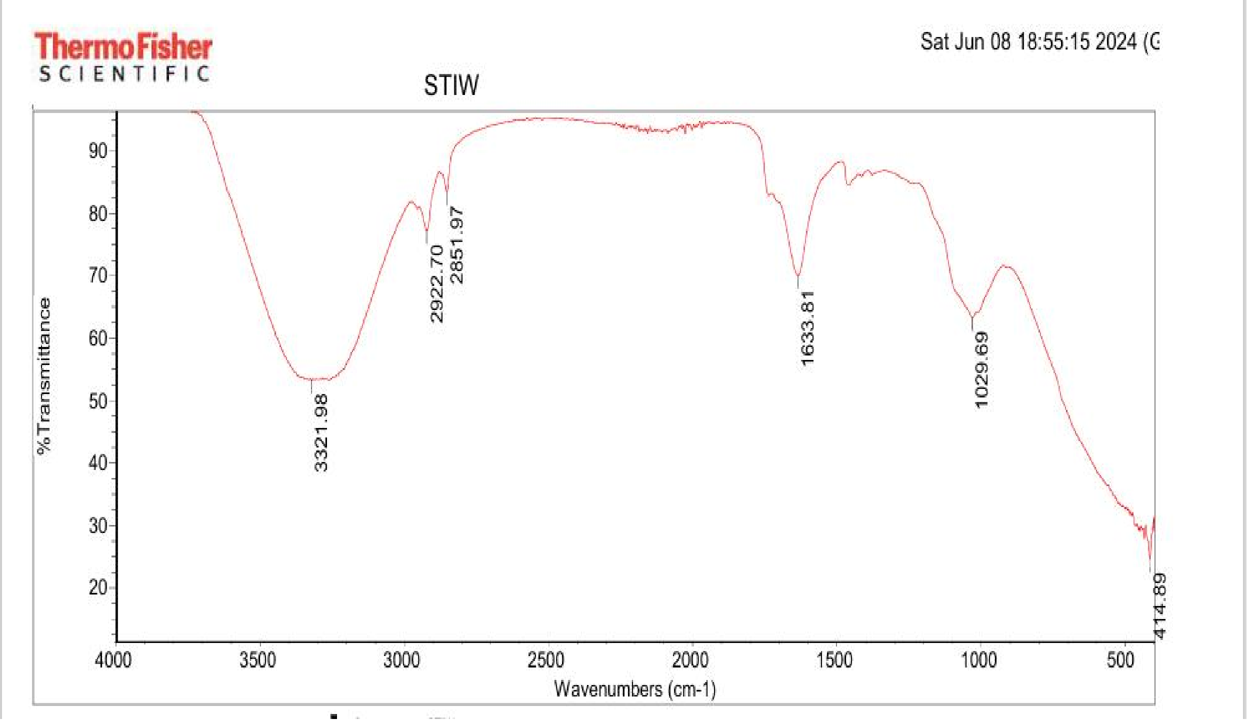

Fig 4.40: FTIR Spectra for Surface Water Sample in Station 1 (March)

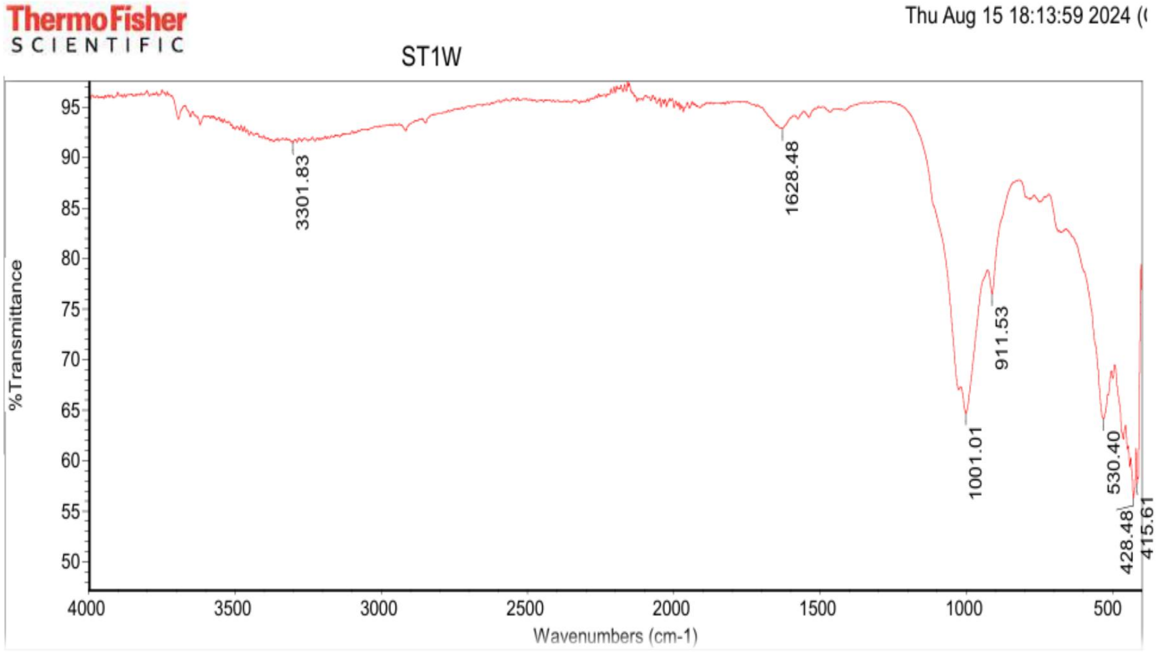

Fig 4.41: FTIR Spectra for Surface Water Sample in Station 1 (May)

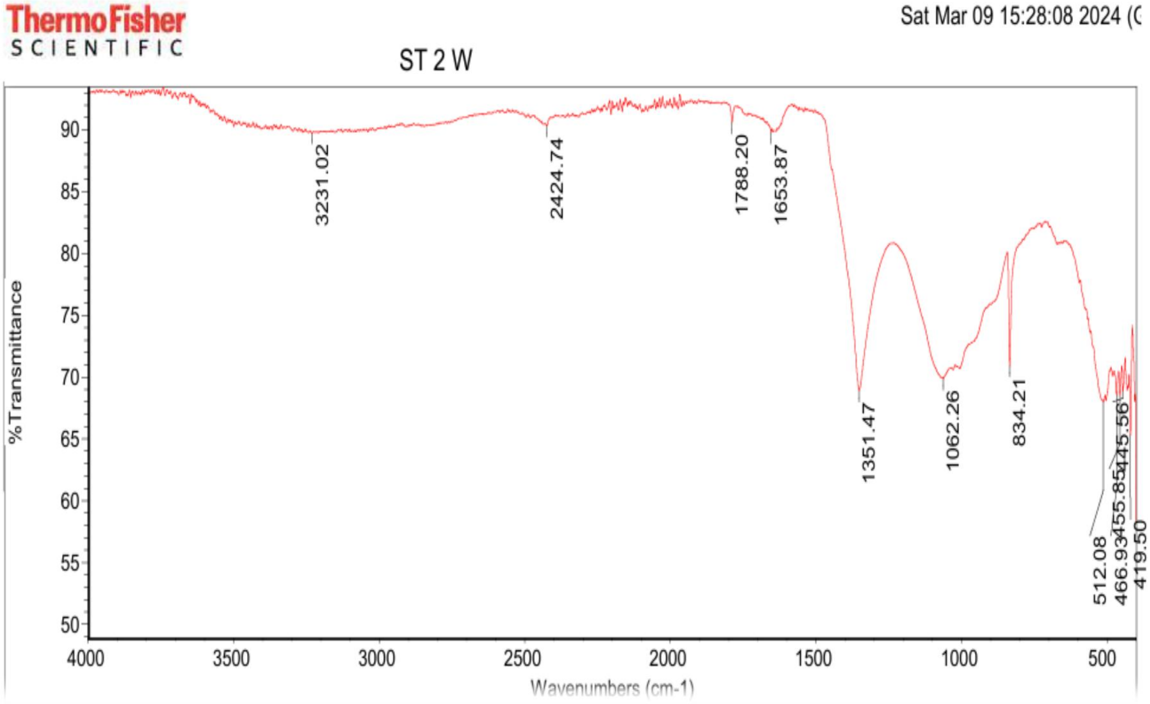

Fig 4.42: FTIR Spectra for Surface Water Sample in Station 2 (January)

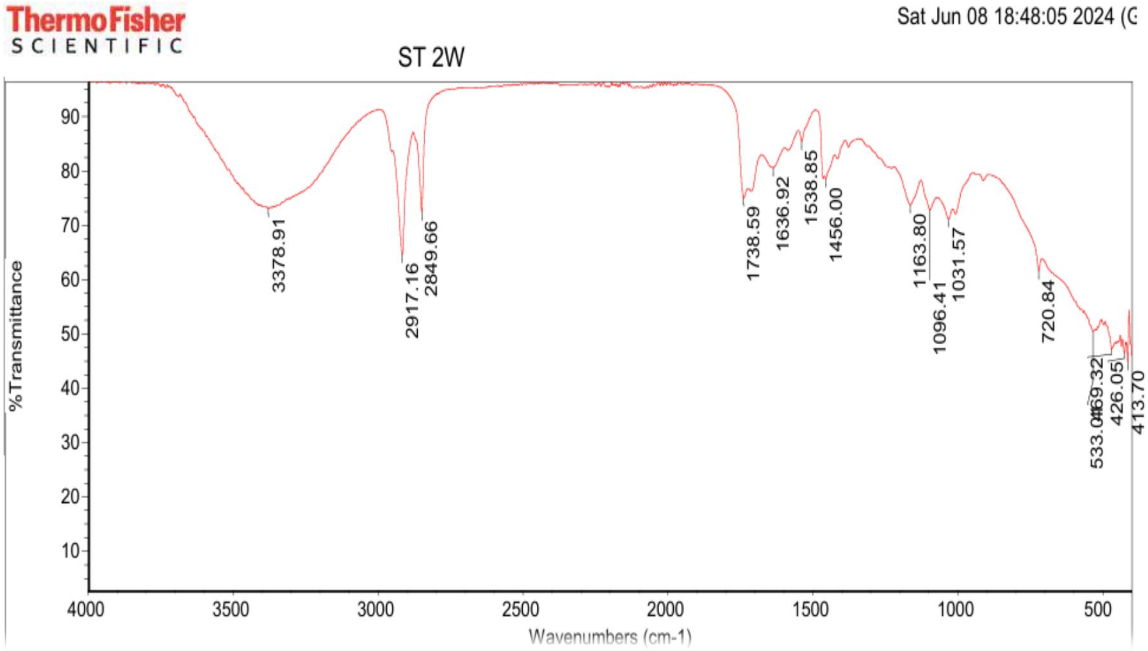

Fig 4.43: FTIR Spectra for Surface Water Sample in Station 2 (March)

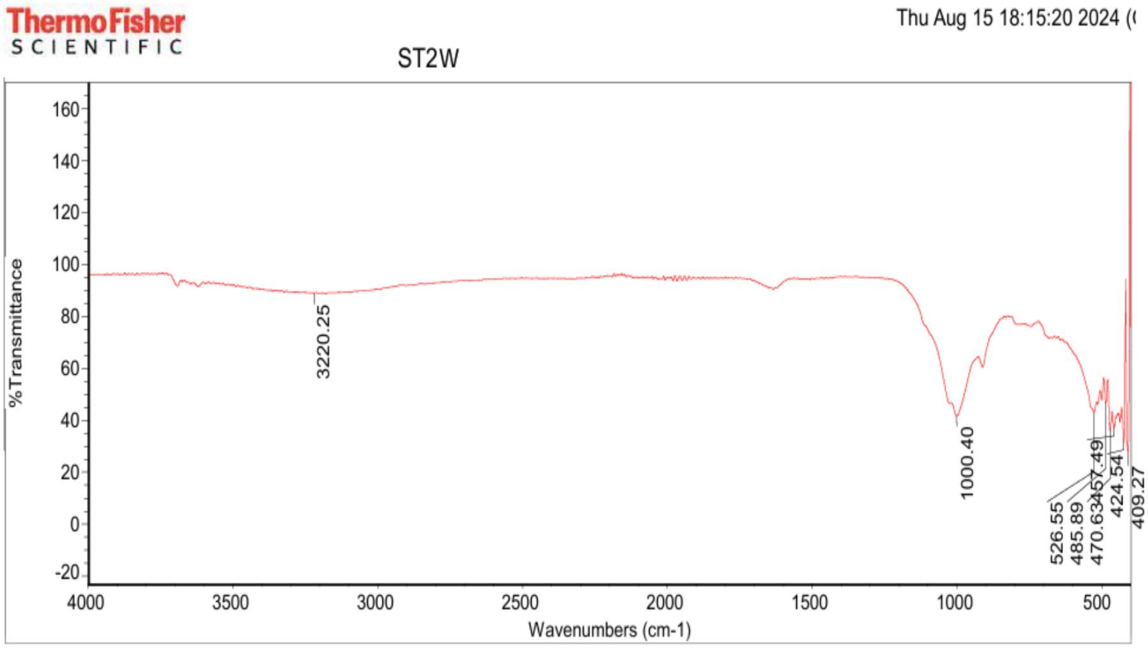

Fig 4.44: FTIR Spectra for Surface Water Sample in Station 2 (May)

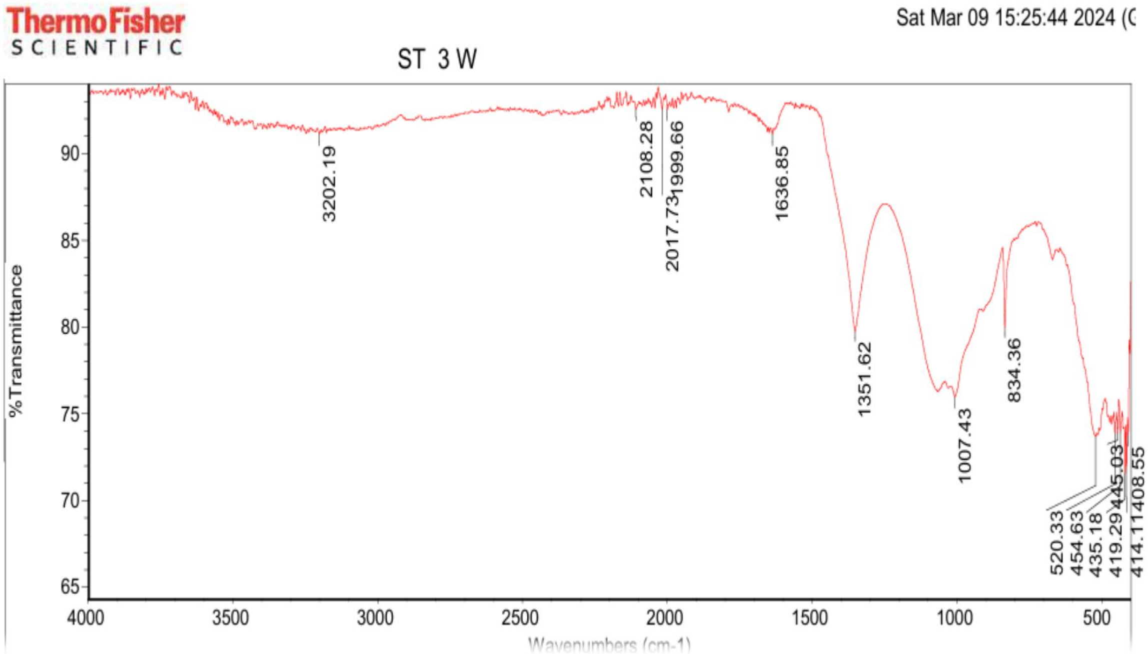

Fig 4.45: FTIR Spectra for Surface Water Sample in Station 3 (January)

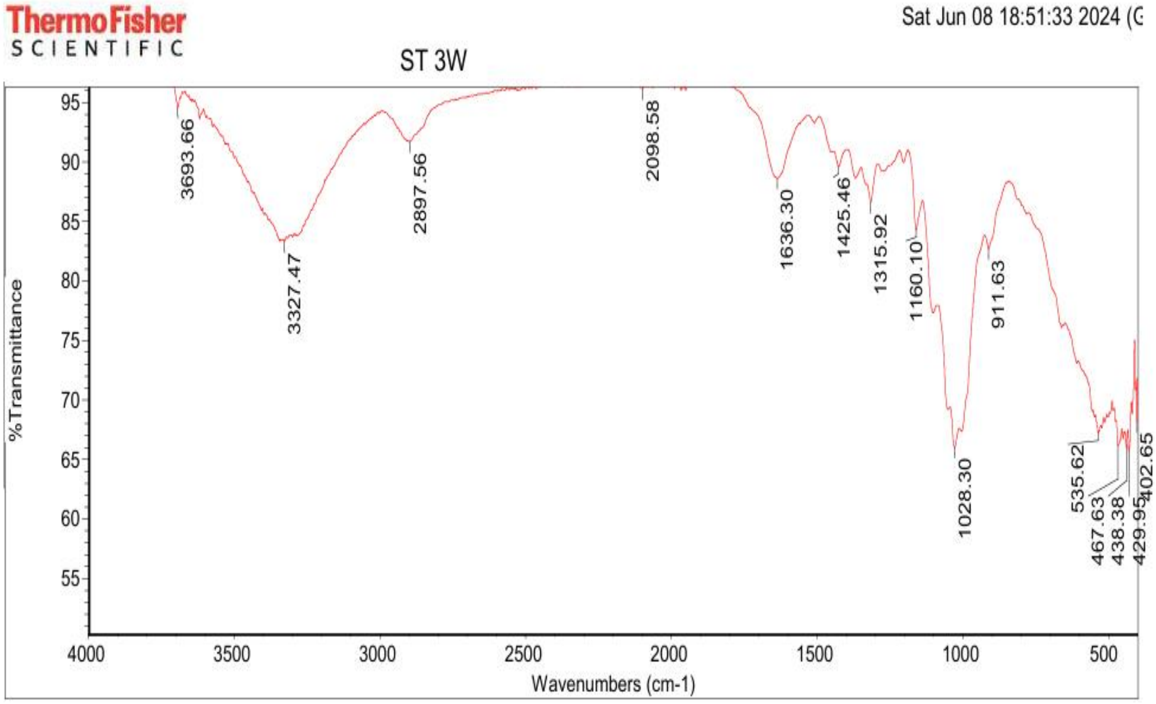

Fig 4.46: FTIR Spectra for Surface Water Sample in Station 3 (March)

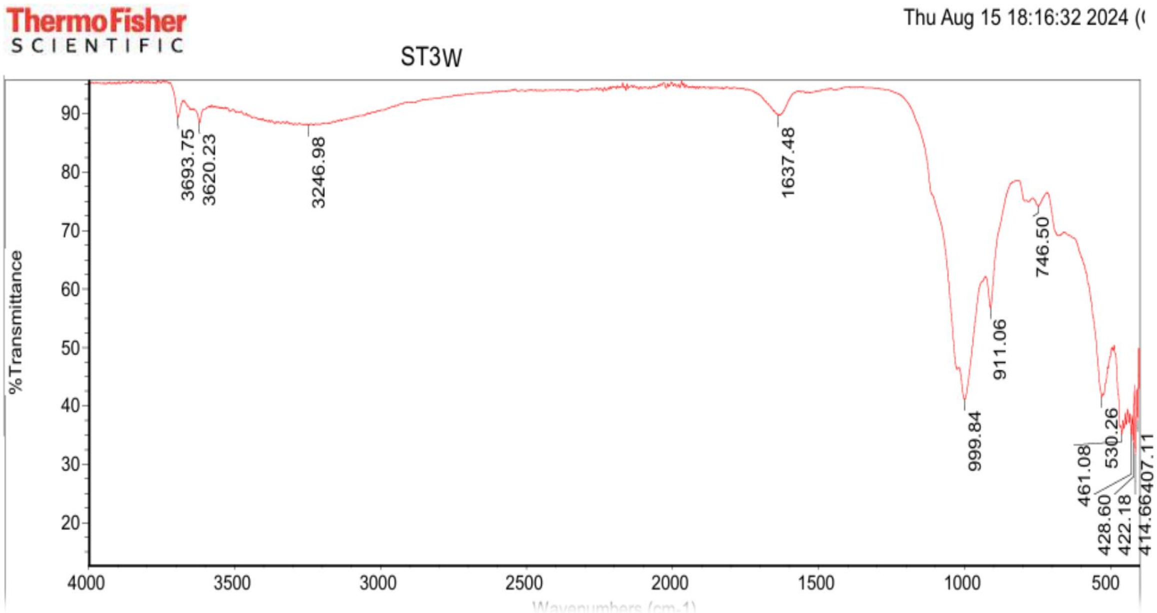

Fig 4.47: FTIR Spectra for Surface Water Sample in Station 3 (May)

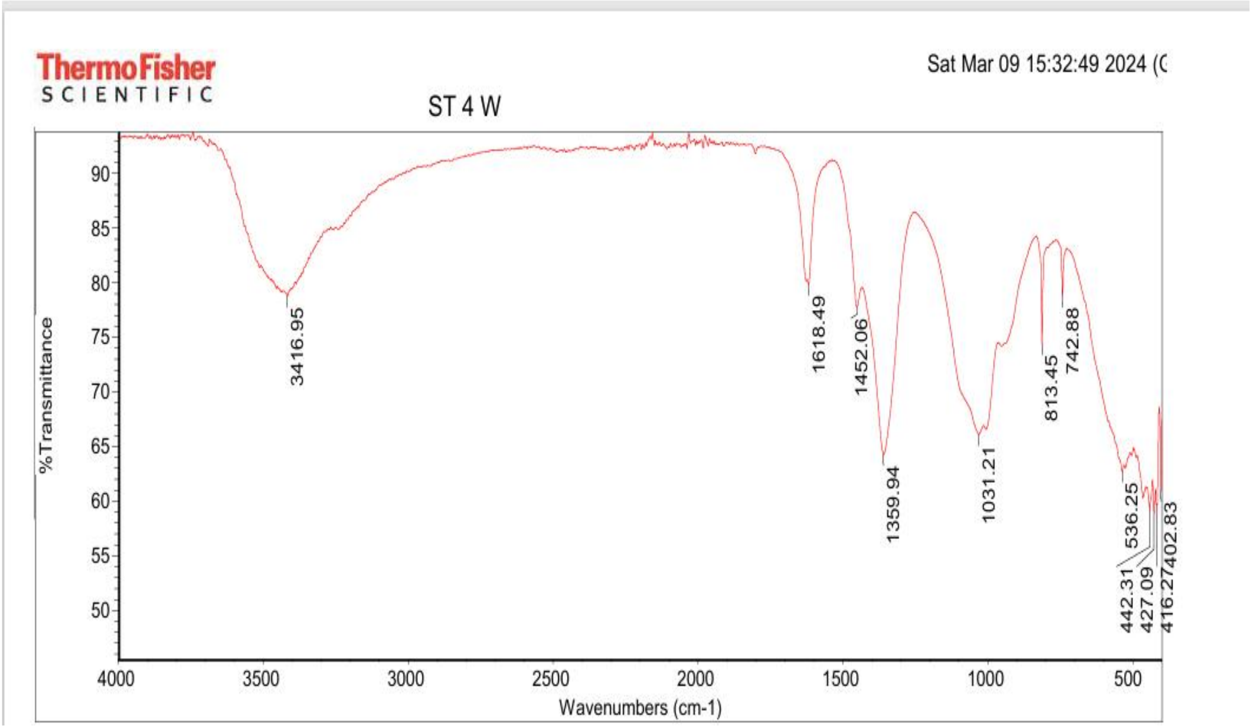

Fig 4.48: FTIR Spectra for Surface Water Sample in Station 4 (January)

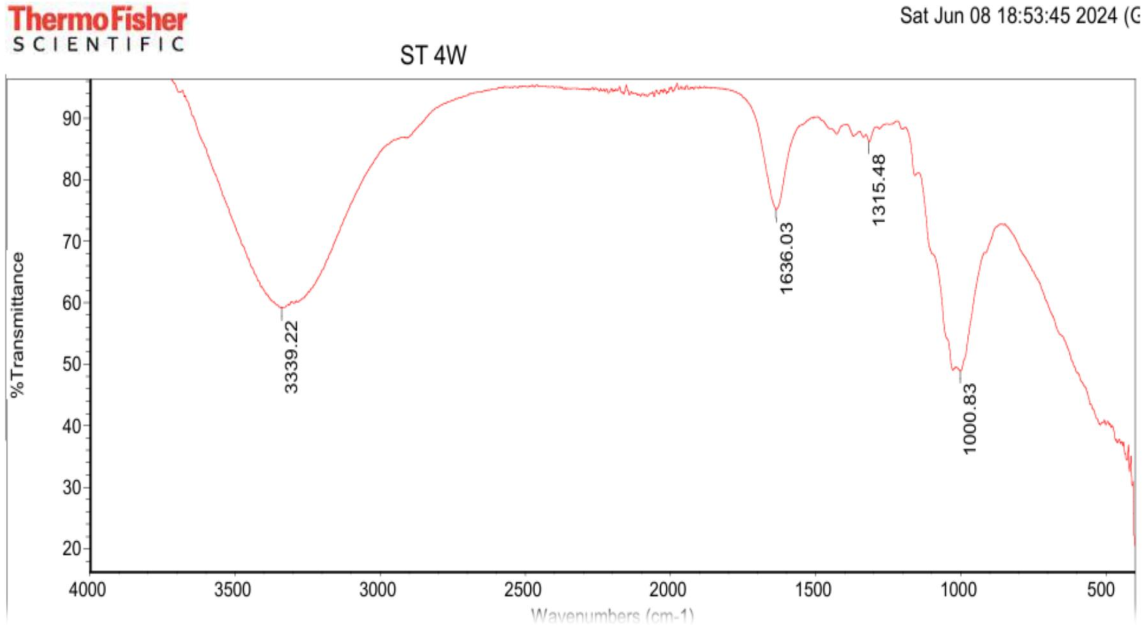

Fig 4.49: FTIR Spectra for Surface Water Sample in Station 4 (March)

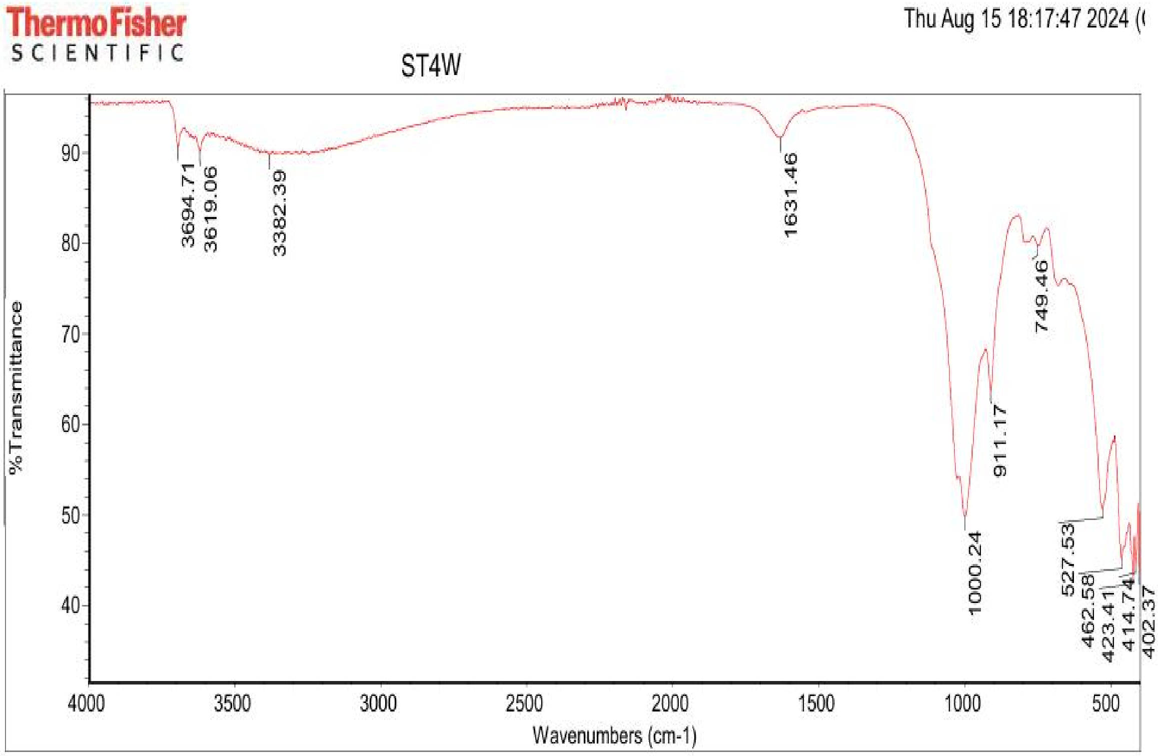

Fig 4.50: FTIR spectra for Water Sample in Station 4 (May)

Table 4.3: FTIR Results by Sampling Station

Station 1

Months	FTIR PEAK (CM⁻¹)	TYPE OF POLYMER	CHARACTERISTIC PEAK	ASSIGNMENT
January	3384.50	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	1636.03	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
March	3321.98	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2922.70	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2851.97	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	1633.81	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
May	3301.83	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	1628.48	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides

Station 2

Months	**FTIR PEAK (CM⁻¹)**	**TYPE OF POLYMER**	**CHARACTERISTIC PEAK**	**ASSIGNMENT**
January	3231.02	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2424.74	Not applicable	C-H stretching	Stretching vibrations indicative of gases or acids
	1788.20	Polyurethane	C=O stretching	Carbonyl stretching indicative of ketones or aldehydes
	1653.87	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
March	3378.91	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2917.16	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2849.66	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	1738.59	Polyethylene Terephthalate (PET)	C=O stretching	Carbonyl stretching indicative of esters
	1636.92	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of proteins
May	3220.25	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups

Station 3

Months	FTIR PEAK (CM^{-1})	TYPE OF POLYMER	CHARACTERISTIC PEAK	ASSIGNMENT
January	3202.19	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2108.28	Polyacetylene	C≡C stretching	Triple bond stretching
	2017.73	Polyacetylene	C≡C stretching	Triple bond stretching
	1999.66	Polyurethane	C=O stretching	Carbonyl stretching
	1636.85	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
March	3693.66	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	3327.47	Cellophane	O-H stretching	Stretching vibrations of -OH groups
	2897.56	High-density Polyethylene (HDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2098.58	Polyacetylene	C≡C stretching	Triple bond stretching
	1636.30	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
May	3693.75	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	3620.23	Cellophane	O-H stretching	Stretching vibrations of -OH groups
	3246.98	Polyethylene Glycol (PEG)	O-H stretching	Stretching vibrations of -OH groups
	1637.48	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides

Station 4

Months	FTIR PEAK (CM⁻¹)	TYPE OF POLYMER	CHARACTERISTIC PEAK	ASSIGNMENT
January	3416.95	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	1618.49	Polyamide (Nylon)	N-H bending / C=O stretching	Bending vibrations of N-H groups or amides
March	3339.22	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	1636.03	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides
May	3694.71	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	3619.06	Cellophane	O-H stretching	Stretching vibrations of -OH groups
	3382.39	Polyethylene Glycol (PEG)	O-H stretching	Stretching vibrations of -OH groups
	1631.46	Polyamide (Nylon)	C=O stretching	Carbonyl stretching indicative of amides

Table 4.4 Summary of Identified Polymer Distributions across Stations

Station	Polyvinyl Alcohol (PVA)	Polyamide (Nylon)	Polypropylene (PP)	Polyethylene (LDPE)	Other Polymers
Station 1	+++	+++	+	+	-
Station 2	+++	++	+	+	Polyurethane (+), PET (+)
Station 3	+++	+++	-	+	PEG (+), High-density Polyethylene (+), Cellophane (++), Polyacetylene (+++)
Station 4	+++	+++	-	-	PEG (+), Cellophane (+)

Note: +++ - High abundant; ++ - Moderate abundant; + - low abundant; - - not detected

4.2.2.1 Polymer Percentage Distribution

At Station 1, Fig 4.51 shows that Polyvinyl Alcohol (PVA) and Polyamide (nylon) were the dominant polymers, each making up 37% of the total microplastics. Polypropylene (PP) and Polyethylene (LDPE) occurred in lower amounts (both at 13%).

Fig 4.52 shows that Polyvinyl Alcohol (PVA) was the most common polymer (34%) at Station 2, followed by Polyamide (nylon (22%). The remaining polymers were Polypropylene (PP), Polyethylene (LDPE), Polyethylene Terephthalate (PET), and Polyurethane, each accounting for 11%.

Fig 4:53 indicates that Polyvinyl Alcohol (PVA) and Polyamide (nylon) each accounted for 22%, making them the dominant polymers. Polyacetylene follows closely at 21%, while Cellophane comprises 14%. High-Density Polyethylene (HDPE**),** Polyethylene Glycol (PEG**),** and Polyurethane each contributed 7%.

At Station 4, Fig 4.54 indicates that Polyvinyl Alcohol (PVA) and Polyamide (nylon**)** each dominate at 37%, while Polyethylene Glycol (PEG) and Cellophane both account for 13%.

Across all four stations as shown in Fig 4.55, Polyvinyl Alcohol (PVA**)** (31%) and Polyamide (nylon) (28%) dominate the overall polymer profile. Polyacetylene and Cellophane each account for 8%, Polypropylene (PP), Polyethylene (LDPE), Polyethylene Glycol (PEG), and Polyurethane each constitute 5%, while High-Density Polyethylene (HDPE) accounts for 3%, and Polyethylene Terephthalate (PET) makes up 2%.

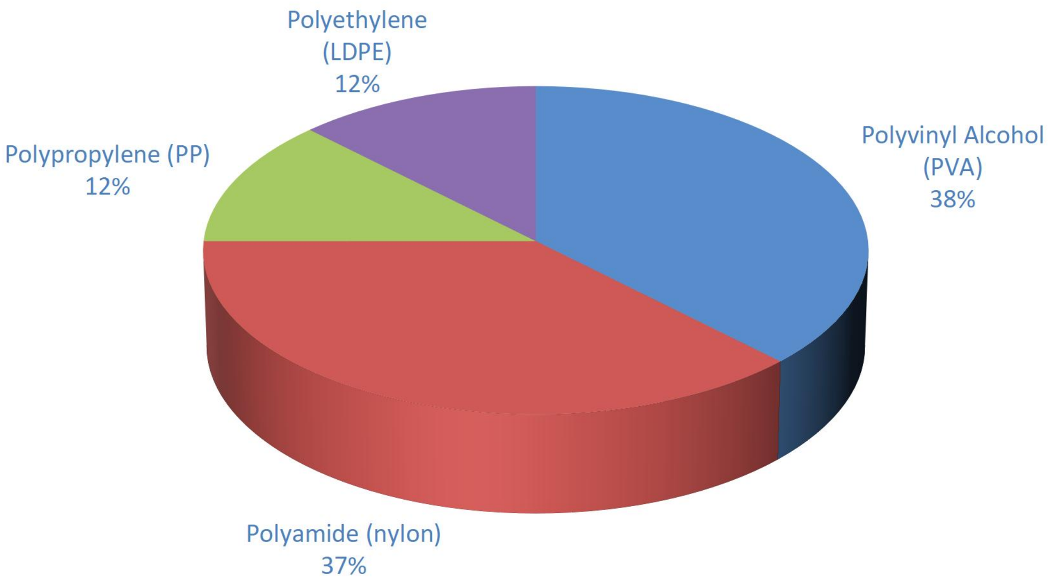

Fig 4.51: Polymer distribution in Station 1

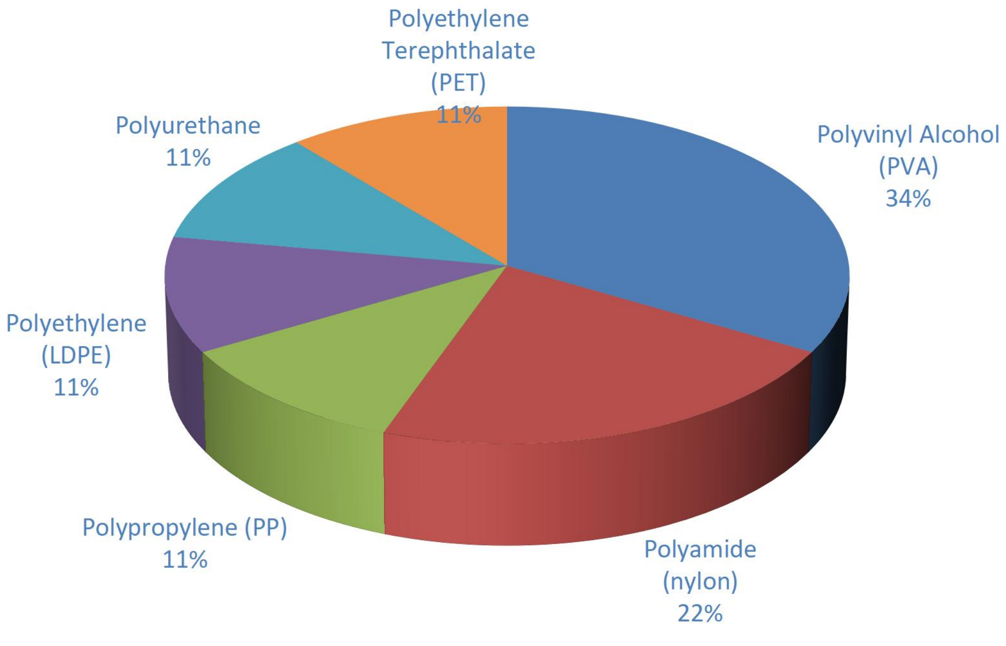

Fig 4.52: Polymer distribution in Station 2

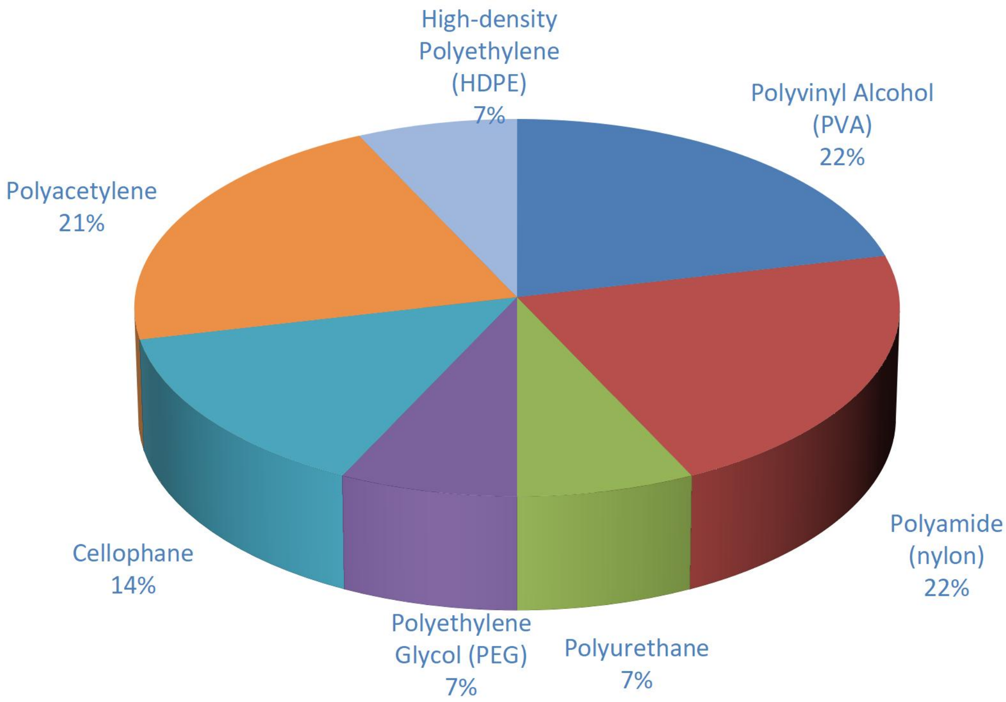

Fig 4.53: Polymer distribution in Station 3

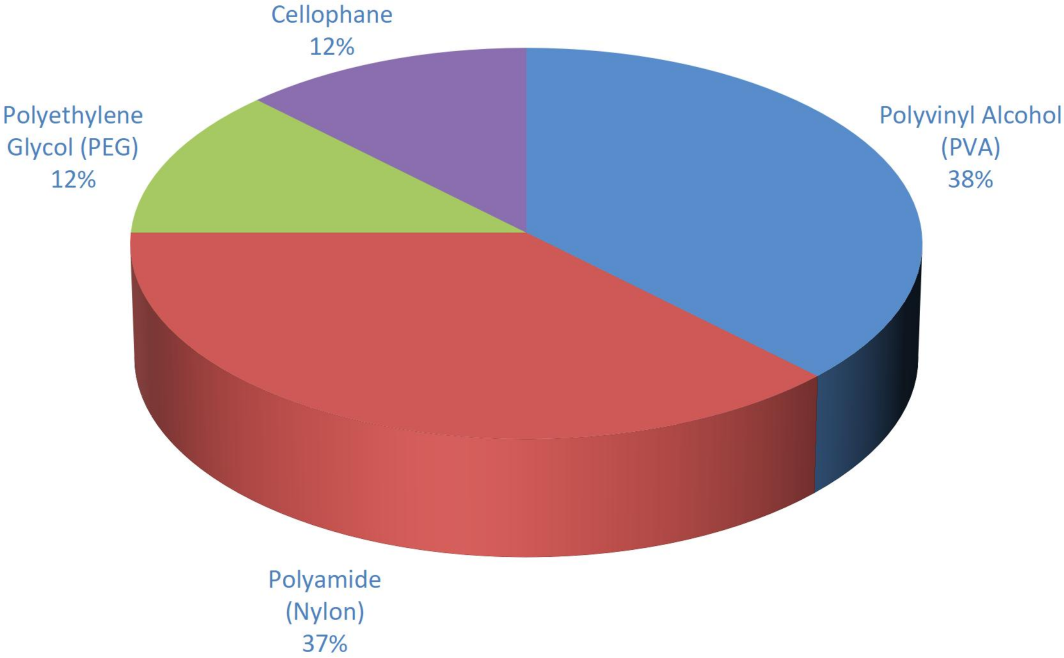

Fig 4.54: Polymer distribution in Station 4

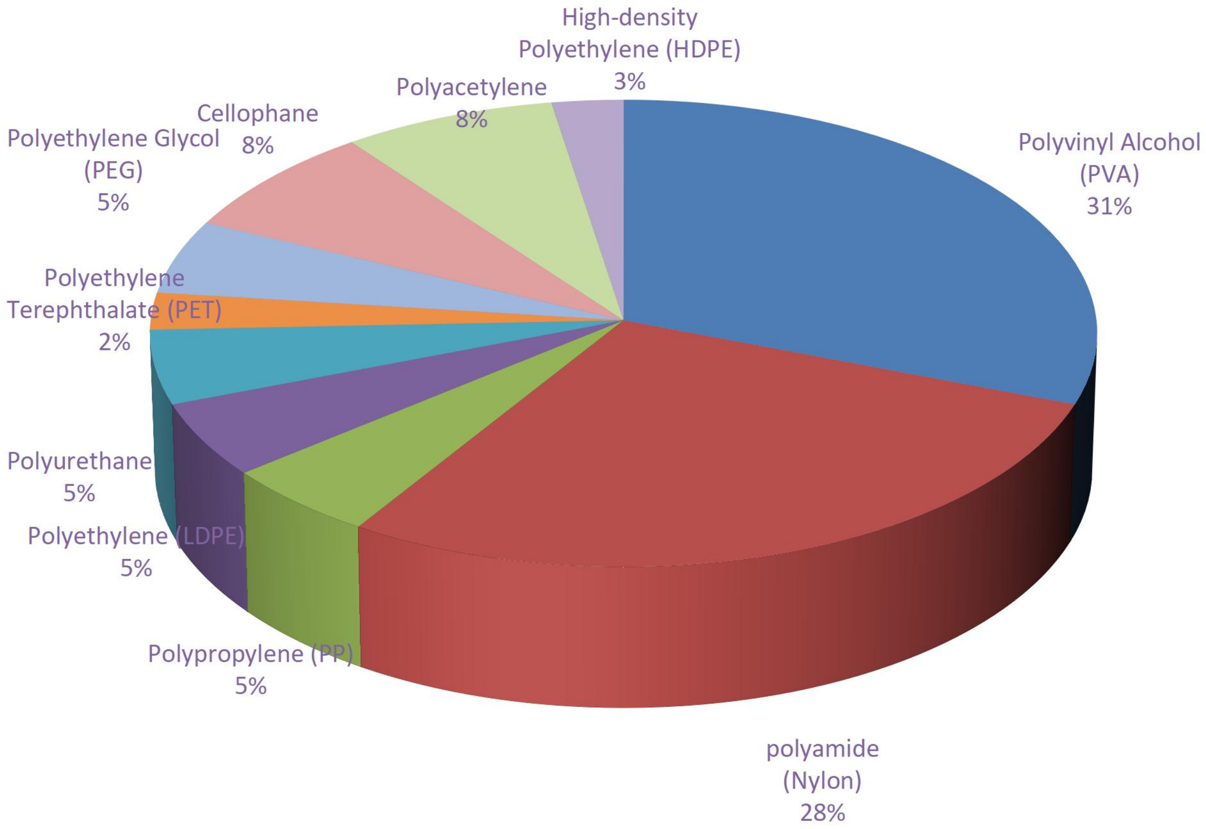

Fig 4.55: Total Polymer Distribution for surface water in the study area

4.2.3 Elemental Composition of Microplastics

The elemental composition of the microplastics based on the Energy Dispersive X-ray Spectroscopy (EDX/EDS) across the study stations is showed in Table 4.5.

At station 1, Chlorine (13.67%), nitrogen (32.87%), sodium (14.25%) and carbon (24%) were recorded in January while carbon (96.02%) dominated in March with minimal contributions from other elements. Sodium (54.48%) and chlorine (36.95%), were highest in May. At station 2, chlorine (9.95%), nitrogen (32.77%), sodium (13.5%) and carbon (28.12%), were recorded in January. In March, aside from noticeable levels of chlorine (6.83%) and sodium (4.74%), a significant spike in silver (43.91%) and sulphur (17.9%) were observed while in May, sodium (16.04%), a pronounced increase in silicon (33.54%), aluminum (14.85%), and iron (4.02%) were recorded. At station 3, moderate chlorine (11.18%), nitrogen (25.68%), sodium (13.79%), and a high carbon content (38.64%) were observed in January, while March samples revealed a dramatic shift with carbon dominating at 88.15% and negligible levels of other elements. By May, the composition shifts again with elevated chlorine (26.77%) and sodium (42.31%), along with increased silicon (8%), aluminum (5.47%), and iron (5.52%). At station 4, chlorine (6.96%) and nitrogen (16.69%) were observed in January, along with high carbon (64.83%). Carbon sharply increased to 91.16% in March, while chlorine and nitrogen dropped significantly. By May, sodium (54.31%) and chlorine (36.92%) dominated, with contributions from silicon (2.92%) and calcium (1.06%).

Table 4.5: Elemental composition of EDX across the four study stations.

Station	Months	Cl (%)	N (%)	Na (%)	C (%)	Ca (%)	Mg (%)	Si (%)	P (%)	K (%)	Al (%)	Fe (%)	S (%)	Ti (%)	Ag (%)	Mn (%)
Station 1	January	13.67	32.87	14.25	24	4.1	3.32	2.73	2.44	1.41	0.96	0.13	0.12	0	--	-
Station 1	March	0.07	3	0.13	96.02	0.02	0.12	0.11	0.22	0.05	0.18	0	0.08	0	-	-
Station 1	May	36.95	-	54.48	-	0.53	0.26	3.12	0.78	0.17	2.78	0.81	0	0.06	-	0.05
Station 2	January	9.95	32.77	13.5	28.12	3.8	1.76	2	4.94	1.22	0.79	0.25	0.88	0	-	-
Station 2	March	6.83	-	4.74	-	2.51	5.64	6.05	3.36	1.43	6.46	0	17.9	1.17	43.91	-
Station 2	May	10.65	-	16.04	-	4.82	1.3	33.54	4.83	8.69	14.85	4.02	0.41	0.3	-	0.54
Station 3	January	11.18	25.68	13.79	38.64	2.21	1.22	1.79	3.3	0.97	0.41	0.21	0.59	0	-	-
Station 3	March	0.37	7.22	1.84	88.15	0.66	0.14	0.48	0.05	0	0.75	0.24	0.07	0.04	-	-
Station 3	May	26.77		42.31	-	2.34	1.32	8	5.94	0.8	5.47	5.52	1.16	0.17	-	0.21
Station 4	January	6.96	16.69	3.7	64.83	3.83	0.97	0.76	1.14	0.68	0.19	0.22	0.04	0	-	-
Station 4	March	0.07	4.91	0	91.16	0.52	0.13	0	0	0	0.12	0	0.09	0	-	-
Station 4	May	36.92	-	54.31	-	1.06	0.67	2.92	1.57	0.35	1.65	0.5	0	0	-	0.05

4.3 Microplastics in Fish

4.3.1 Particle Size Distribution (PSD) Analysis

The particle size distribution (PSD) of microplastics in fish species are presented in Table 4.6. In January, *Notopterus afer.* and *Heterobranchus bidorsalis* recorded their highest mean PSDs at 1.55 ± 0.311 µm and 1.50 ± 0.50 µm, respectively. Notably, *Notopterus afer* exhibited slightly higher PSD values with lower variability. By March, mean PSD values had declined in both species, with *Notopterus afer* at 1.00 ± 0.50 µm and *Heterobranchus bidorsalis* at 1.04 ± 0.77 µm. While in May, both species exhibited the lowest mean PSDs: 0.56 ± 0.78 µm for *Notopterus afer* and 0.53 ± 0.57 µm for *Heterobranchus bidorsalis* Fig 4.56-4.61 illustrated the particle size distribution of microplastics identified in fish samples.

Table 4.6: Summary of PSD (µm) in Fish

Fish Species	January (Mean ± SD)	March (Mean ± SD)	May (Mean ± SD)
Notopterus afer	1.55 ± 0.31	1.00 ± 0.50	0.56 ± 0.78
Heterobranchus bidorsalis.	1.50 ± 0.50	1.04 ± 0.77	0.53 ± 0.57

SAMPLE: A

PARTICLE SIZE DISTRIBUTION

1

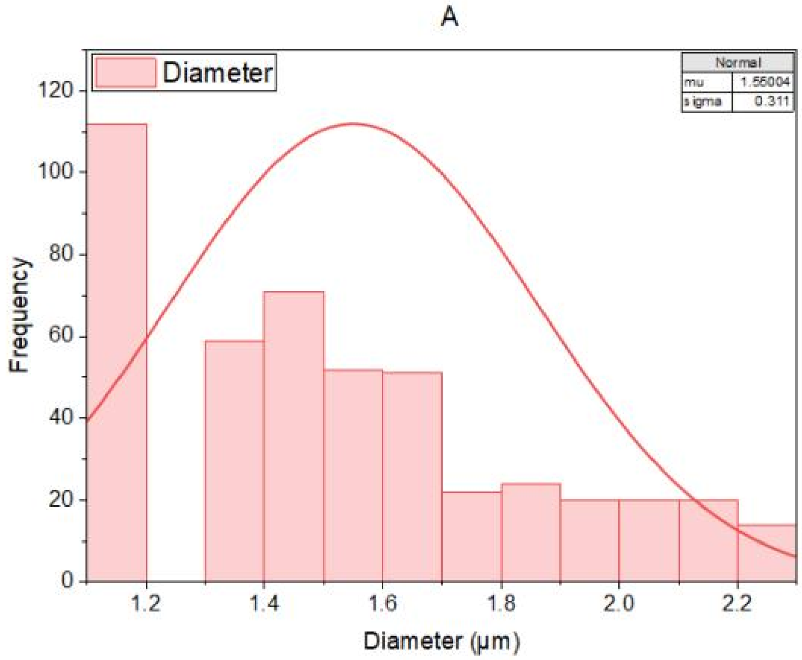

Fig 4.56: Particle Size Distribution of *Notopterus afer* for January

SAMPLE: E

PARTICLE SIZE DISTRIBUTION

1

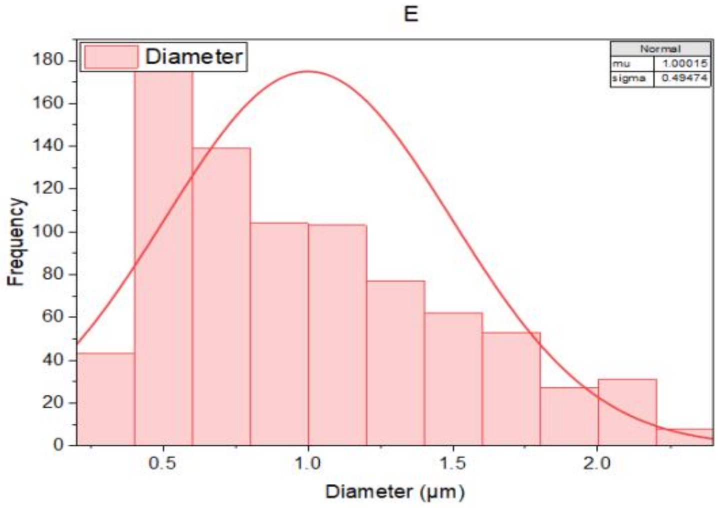

Fig 4.57: Particle Size Distribution of *Notopterus afer* for March

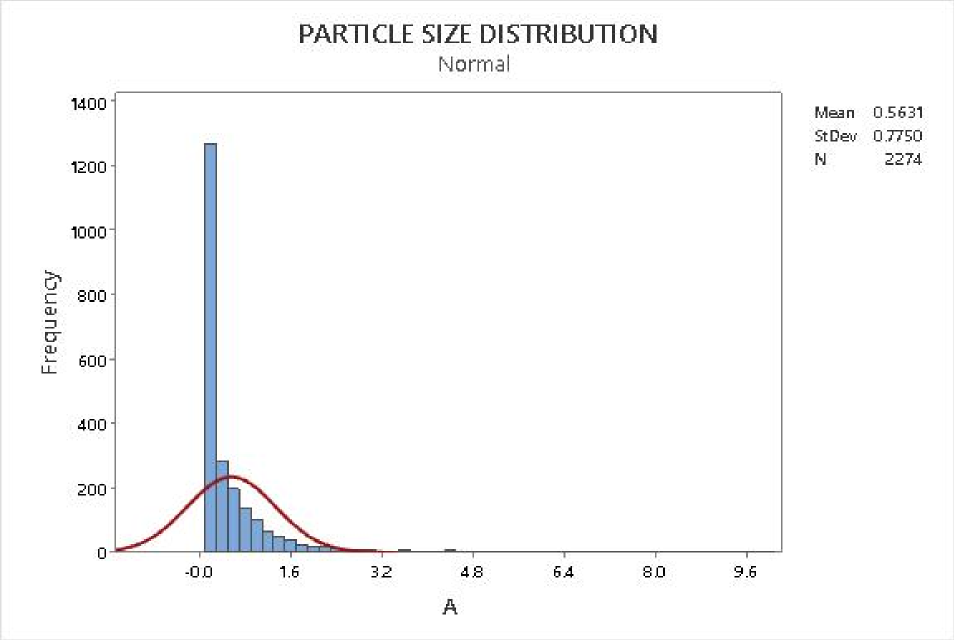

Fig 4.58: Particle Size Distribution of *Notopterus afer* for May

SAMPLE: B

PARTICLE SIZE DISTRIBUTION

1

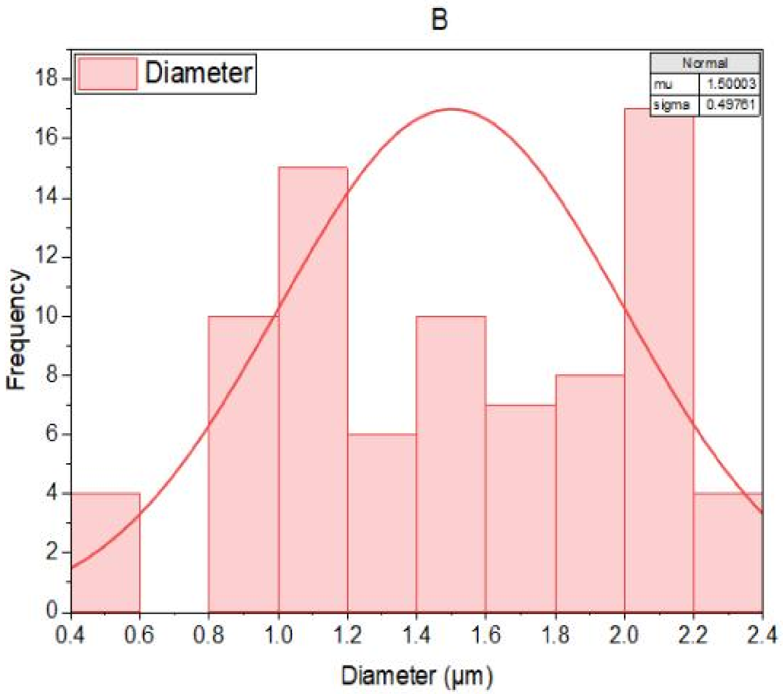

Fig 4.59: Particle Size Distribution of *Heterobranchus bidorsalis* for January

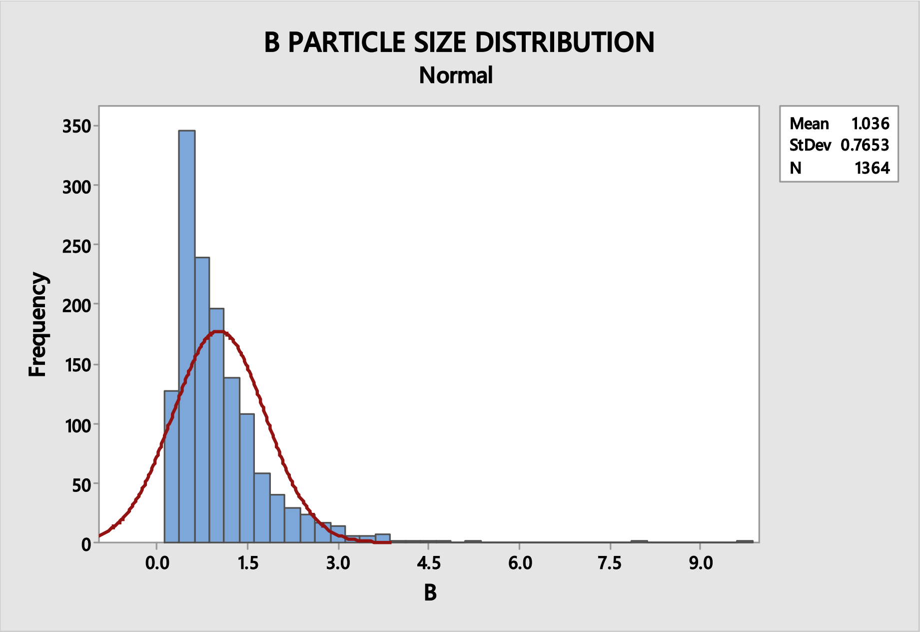

Fig 4.60: Particle Size Distribution of *Heterobranchus bidorsalis* for March

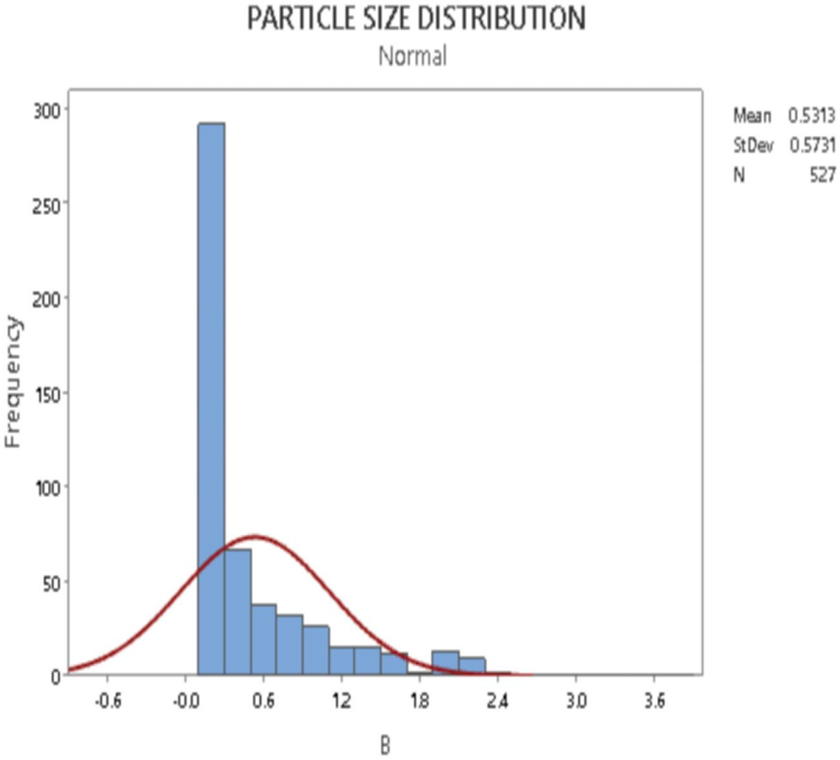

Fig 4.61: Particle Size Distribution of *Heterobranchus bidorsalis* for May

4.3.2 Polymer Analysis of Microplastics in Fish

Fourier-transform infrared spectroscopy (FTIR) analysis of fish samples revealed the presence of five distinct polymers: polypropylene (PP), low-density polyethylene (LDPE), polyethylene terephthalate (PET), polyvinyl alcohol (PVA), and cellophane. The results indicate that *Notopterus afer* ingested four polymer types, PP, LDPE, PET, and PVA while *Heterobranchus bidorsalis* contained all five, including cellophane. The characteristic FTIR peaks used for polymer identification include, CH_2 and CH_3 stretching (2850–2922 cm^{-1}) associated with PP and LDPE, C=O stretching (around 1744 cm^{-1}) characteristic of PET, and O–H stretching (3370–3700 cm^{-1}) indicative of PVA and cellophane. Table 4.7 summarizes the FTIR peaks observed from both species across the three sampling periods while FTIR spectra were used to identify the functional group region as presented in Fig 4.62-4.67

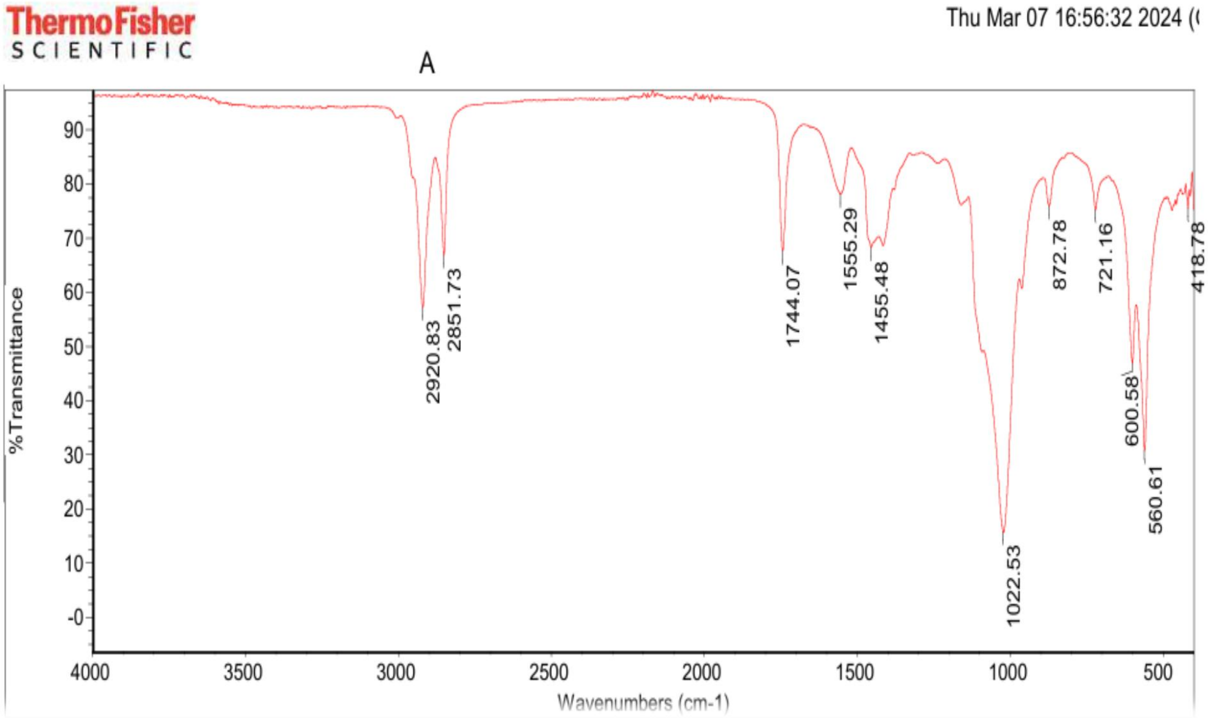

Fig 4.62: FTIR spectra for *Notopterus afer* (January)

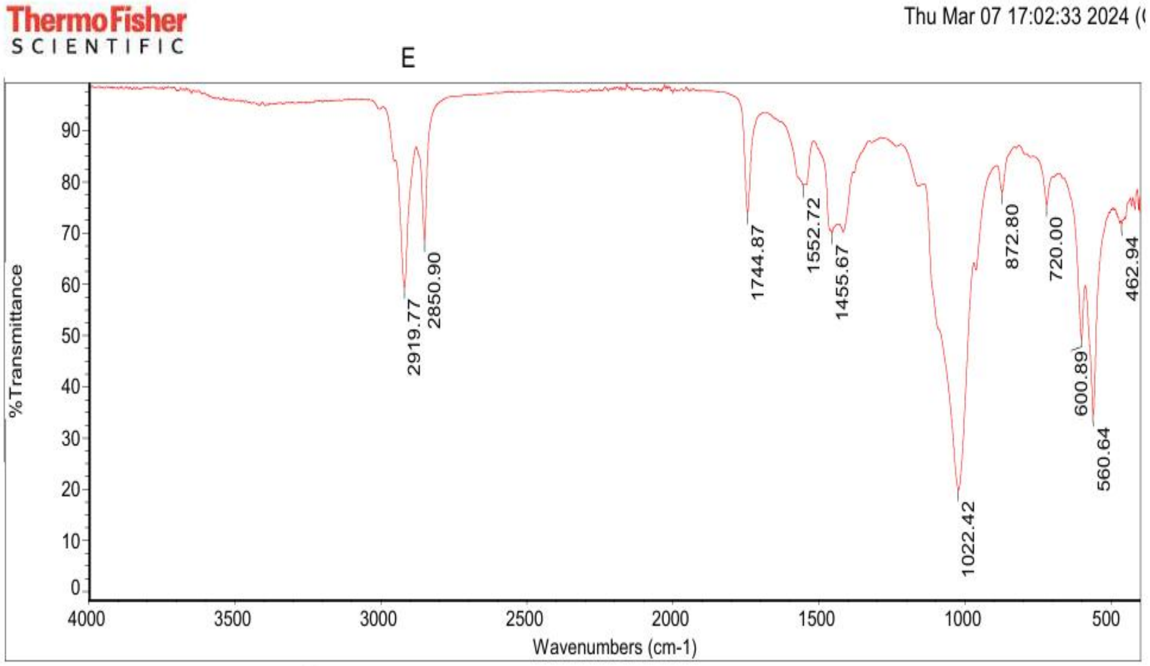

Fig 4.63: FTIR spectra for *Notopterus afer* (March)

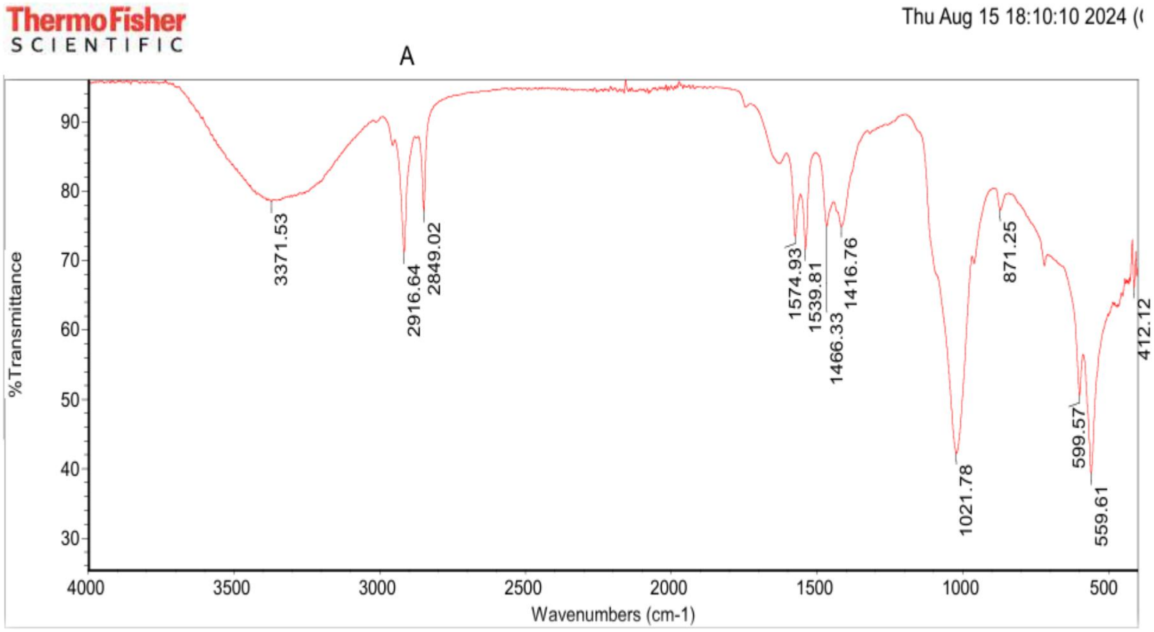

Fig 4.64: FTIR spectra for *Notopterus afer* (May)

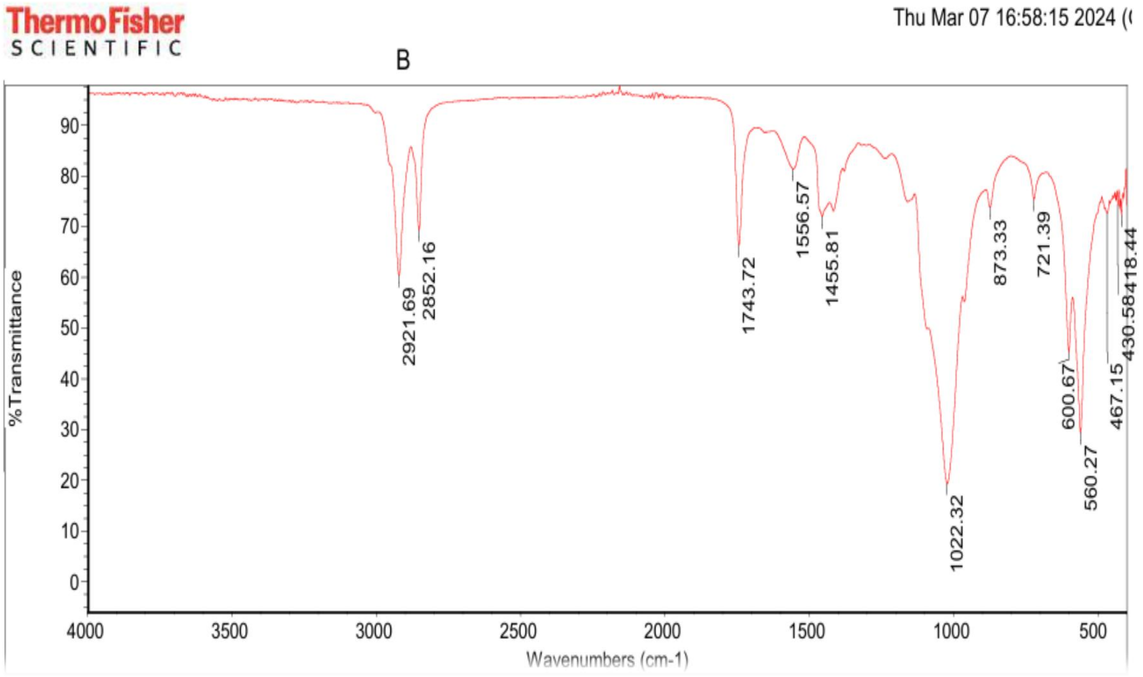

Fig 4.65: FTIR spectra for *Heterobranchus bidorsalis* (January)

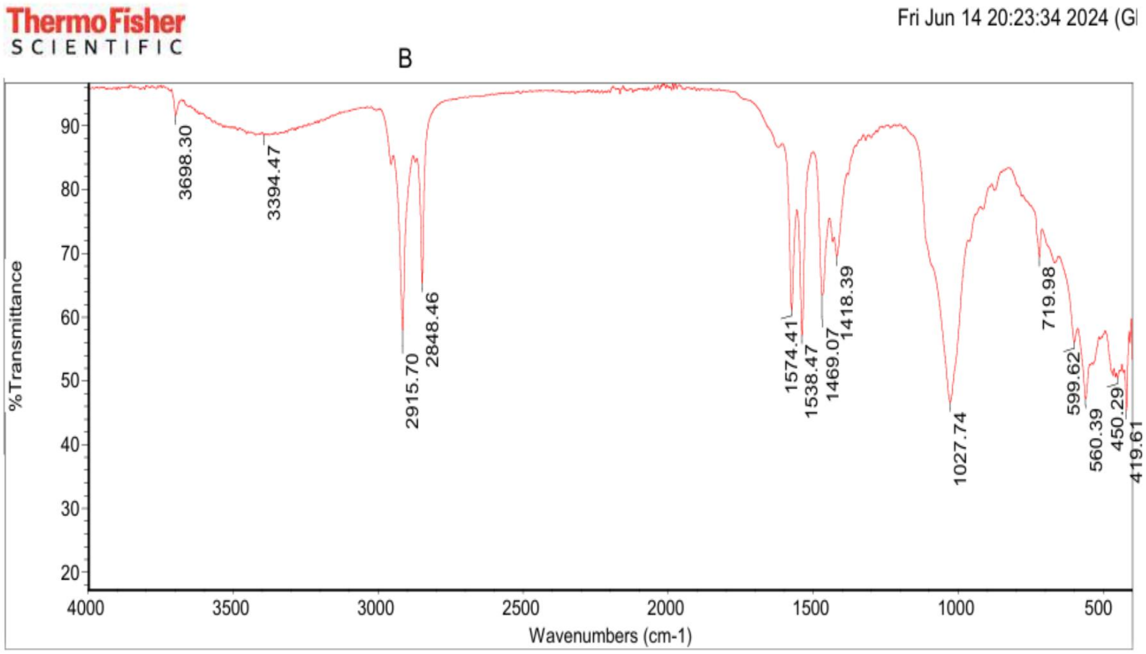

Fig 4.66: FTIR spectra for *Heterobranchus bidorsalis* (March)

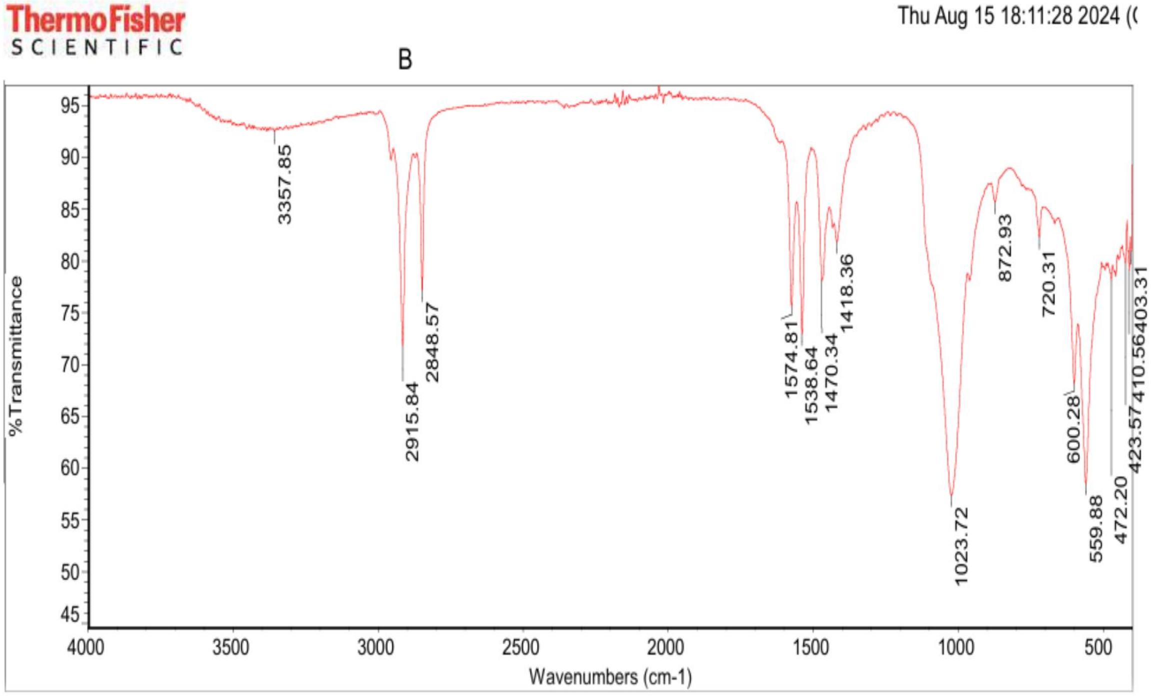

Fig 4.67: FTIR Spectra for *Heterobranchus bidorsalis* (May)

Table 4.7: Summary of FTIR Peaks

Notopterus afer

MONTHS	FTIR PEAK (CM^{-1})	TYPE OF POLYMER	CHARACTERISTIC PEAK	ASSIGNMENT
January	2920.83	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2851.73	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	1744.07	Polyethylene Terephthalate (PET)	C=O stretching	Carbonyl stretching in esters
March	2919.77	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2850.90	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	1744.87	Polyethylene Terephthalate (PET)	C=O stretching	Carbonyl stretching in esters
May	3371.53	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2916.64	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2849.02	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups

Heterobranchus bidorsalis

SAMPLING TIME	FTIR PEAK (CM^{-1})	TYPE OF POLYMER	CHARACTERISTIC PEAK	ASSIGNMENT
January	2921.69	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2852.16	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	1743.72	Polyethylene Terephthalate (PET)	C=O stretching	Carbonyl stretching in esters
March	3698.30	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	3394.47	Cellophane	O-H stretching	Stretching vibrations of -OH groups
	2915.70	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2848.46	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
May	3357.85	Polyvinyl Alcohol (PVA)	O-H stretching	Stretching vibrations of -OH groups
	2915.84	Polypropylene (PP)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups
	2848.57	Polyethylene (LDPE)	CH2 and CH3 stretching	Stretching vibrations of methylene and methyl groups

Table 4.8 Summary of Identified Polymers Distribution Among the Fish Species

Fish *Spps*	Polypropylene (PP)	Polyethylene (LDPE)	Polyethylene Terephthalate (PET)	Polyvinyl Alcohol (PVA)	Other Polymers
Notopterus afer	+++	+++	++	+	-
Heterobranchus bidorsalis	+++	+++	+	++	Cellophane (+)

Note: +++ - High abundant; ++ - Moderate abundant; + - low abundant; - - not detected

4.3.2.1 Polymer Percentage Distribution

The dominant polymer type in *Notopterus afer* as shown in Fig 4.68 is Polypropylene (PP) (34%), followed closely by Polyethylene (LDPE) (33%), Polyethylene Terephthalate (PET) (22%) and Polyvinyl Alcohol (PVA) (11%). In *Heterobranchus bidorsalis* (Fig 4.69), the microplastic profile was dominated by Polypropylene (30%) and Polyethylene (LDPE) (30%), followed by Polyvinyl Alcohol (PVA) (20%), Polyethylene Terephthalate (PET) (10%), and Cellophane (10%). Across both fish species as shown in Fig 4.70 were Polypropylene (31%) and Polyethylene (LDPE) (32%). Other polymers include; Polyvinyl Alcohol (PVA) (16%), Polyethylene Terephthalate (PET) (16%), and Cellophane (5%).

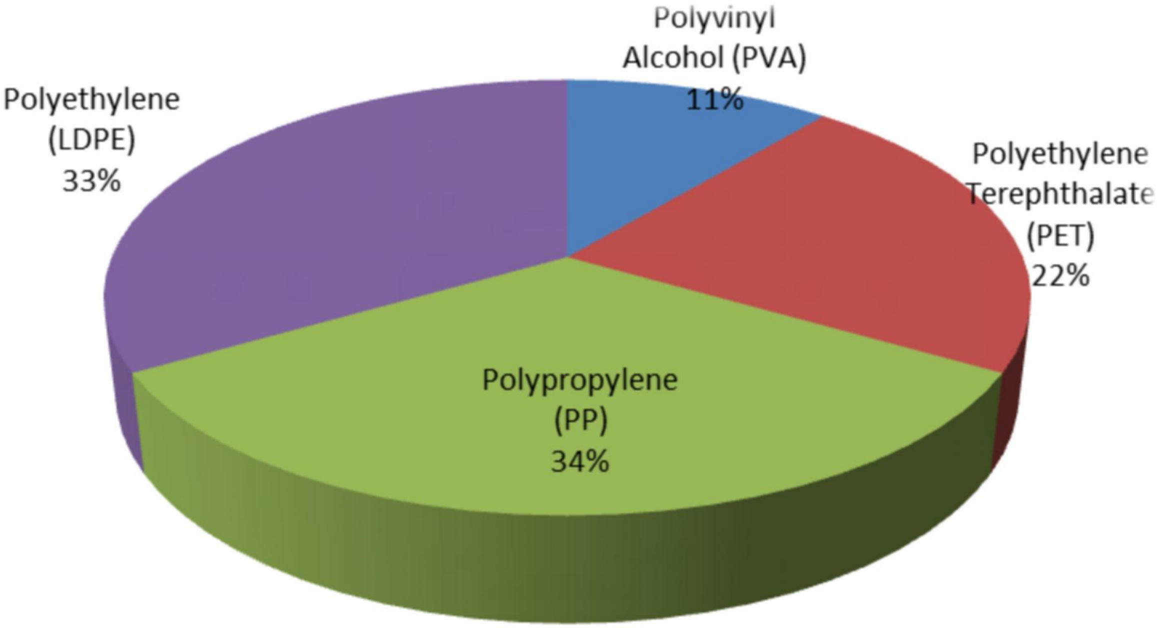

Fig 4.68: Polymer distribution in *Notopterus afer*

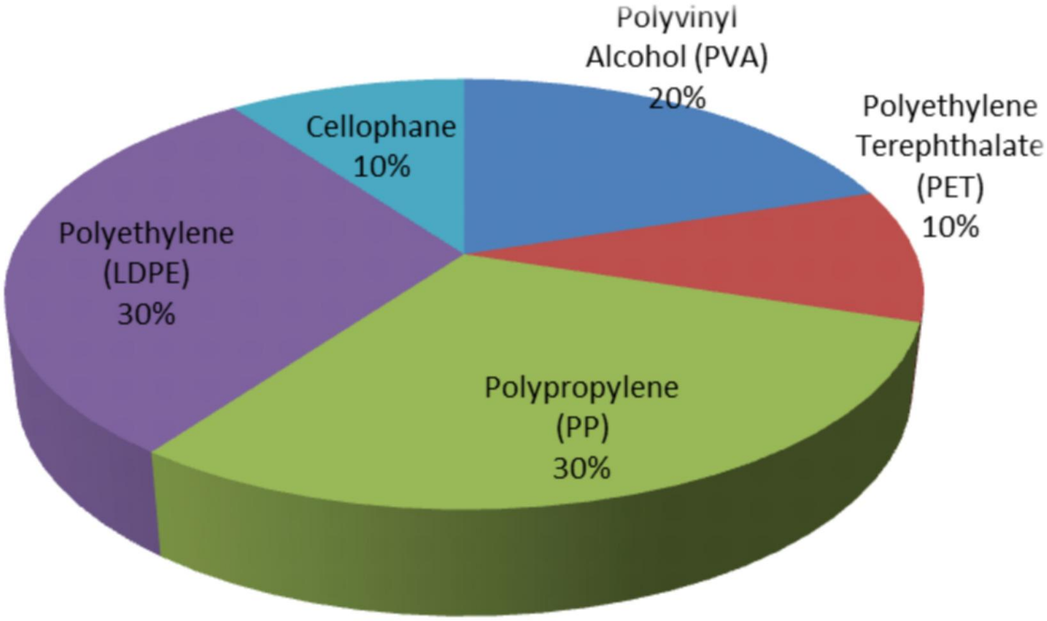

Fig 4.69: Polymer distribution in *Heterobranchus bidorsalis*

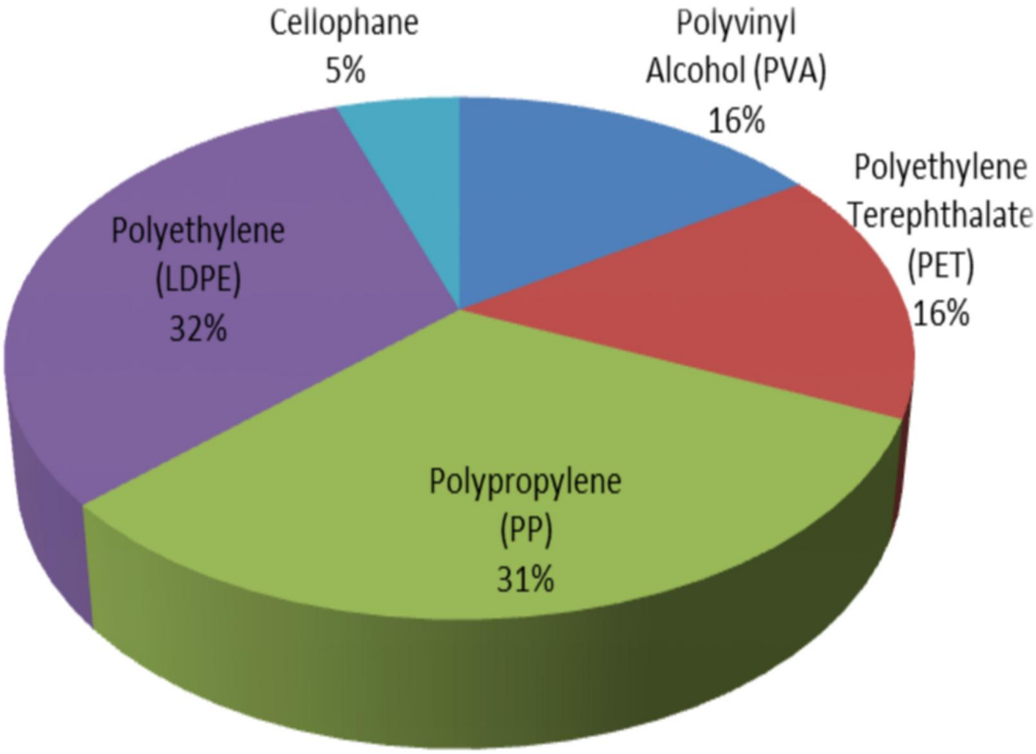

Fig 4.70: Total Polymer Distribution for fish in the study area

4.3.3 Elemental Composition of Microplastic in Fish of Ovia River

The elemental composition of microplastics extracted from *Notopterus afer* and *Heterobranchus bidorsalis* during the sampling periods is presented in Table 4.9. In *Notopterus afer*, the January samples were dominated by carbon, with other elements including nitrogen, chlorine, sodium, silicon, and calcium. By March, carbon levels remained stable at 80.68%, while nitrogen and chlorine increased to 6.52% and 3.09%, respectively. Sodium was present at 1.69%, along with minor concentrations of calcium (1.06%) and phosphorus (2.37%). A notable shift occurred in May, where calcium and phosphorus became the predominant elements. Sodium also increased to 8.88%, while carbon declined significantly. Other elements identified in May include magnesium (1.25%), sulfur (1.13%), potassium (1.33%), and aluminum (1.15%).

In *Heterobranchus bidorsalis*, January samples showed a high carbon concentration, with low levels of nitrogen, chlorine, silicon, sodium, and calcium. In March, there was a noticeable decline in carbon, with sodium emerging as the dominant element, followed by calcium, phosphorus, chlorine, potassium, and aluminum. By May, calcium and phosphorus dominated the composition, with sodium increasing to 6.39%. Carbon continued to decline, and additional elements such as magnesium (1.45%), sulfur (1.16%), and titanium (0.76%) were detected in trace amounts.

Table 4.9: Summary of Elemental Composition of EDX for Fish Species

Fish	Sampling Time	Cl (%)	N (%)	Na (%)	C (%)	Ca (%)	Mg (%)	Si (%)	P (%)	K (%)	Al (%)	Fe (%)	S (%)	Ti (%)
Notopterus afer	January	0.38	2	0.42	80.58	4.23	0.43	0.73	1.83	0.17	0.12	0.08	0.02	0
	March	3.09	6.52	2.2	80.68	4.08	0.63	0.22	1.52	1.06	0	0	0	0
	May	0.24		8.88		51.29	3.56	5.96	26.93	1.74	1.23	0.17	0	0
Heterobranchus bidorsalis	January	0.04	3.04	0.49	91.73	2.66	0.21	0.28	1.46	0.15	0.05	0.05	0	0.04
	March	1.38	3.2	8.34		0.75	1.22	2.43	1.22	0	83.06	0.35	1.25	0
	May	0.18		1.52		60.72	4.29	1.52	26.9	3.02	1.24	0.6	0	0

4.4 Water Quality Index (WQI) of study area

The calculated WQI values for the sampling stations indicate varying water quality levels**.** Station 1 recorded a WQI of 47.71**,** classifying it as good water quality. Station 2 had a WQI of 72.76, indicating poor water quality**.** Station 3 exhibited the highest WQI at 90.29**,** representing very poor water quality**.** Station 4 recorded a WQI of 64.60**,** also falling under the poor water quality category. The results highlight a decline in water quality across most stations, with only Station 1 maintaining good conditions. The pollution status classification for each station is summarized in Table 4.10.

Table 4.10: Overall Water Quality Index (WQI) for all stations and the overall result:

Station	WQI Value	Pollution Status
Station 1	47.71	Good Water Quality
Station 2	72.76	Poor Water Quality
Station 3	90.29	Very Poor Water Quality
Station 4	64.60	Poor Water Quality

4.2 Comprehensive Pollution Index

The Pollution Index (PI) results presented in Table 4.12 evaluate pollution levels for each water quality parameter across stations, using regulatory standards as reference. A PI greater than 1 indicates pollution. For pH, PI values across all stations range from 0.79 to 0.81, indicating slight pollution despite values being within the regulatory range (6.5–8.5). Electrical conductivity (EC) had consistently low PI values (0.08), suggesting no pollution.

Turbidity showed medium pollution across stations, with PI values between 1.21 and 1.70 (Table 4.12), while dissolved oxygen (DO) values also indicated medium pollution with PI values from 1.27 to 1.33, despite concentrations being above 5 mg/L. Biological oxygen demand (BOD) presented slight pollution, with PI values ranging from 0.56 to 0.60 (Table 4.12). Nitrate and phosphate had very low PI values (0.05 and 0.28), indicating non-pollution. Iron showed slight pollution, with PI values between 0.47 and 0.87. Lead displayed variable pollution levels: Stations 1 and 3 had PI values of 1.00 (slight pollution), while Stations 2 and 4 recorded 0.00 (non-pollution).

Table 4.13 provides the actual measured values and pollution grades for each parameter. pH (6.73–6.87) confirms slight pollution across stations, while EC values are far below standard thresholds, confirming non-pollution. Turbidity values exceed the 5 NTU limit, supporting the medium pollution classification. DO, nitrate, and phosphate levels align with the PI findings. Iron and lead also follow the same pollution pattern slight to none depending on station.

The Composite Pollution Index (CPI) summarized in Table 4.14 shows all stations fall under Level III (slight pollution). Station 3 recorded the highest CPI (0.74), followed by Station 1 (0.69), Station 4 (0.59), and Station 2 (0.53).

Table 4.14: Overall CPI Values and Pollution Status of All Stations

Station	CPI	Water Quality Level	Pollution Status
Station 1	0.69	Level III	Slight pollution
Station 2	0.53	Level III	Slight pollution
Station 3	0.74	Level III	Slight pollution
Station 4	0.59	Level III	Slight pollution

Table 4.12: Pollution Index (PI) Parameters across All Stations

Parameter	Regulatory Standard (Si)	Station 1 PI	Station 2 PI	Station 3 PI	Station 4 PI	Pollution Grade
pH	6.5–8.5	0.79	0.79	0.81	0.81	Slight pollution
Electrical Conductivity (EC)	1000 µS/cm	0.08	0.08	0.08	0.08	Non-pollution
Turbidity	5 NTU	1.66	1.21	1.70	1.42	Medium pollution
Dissolved Oxygen (DO)	5 mg/L	1.31	1.32	1.27	1.33	Medium pollution
BOD	6 mg/L	0.60	0.58	0.56	0.59	Slight pollution
Nitrate	50 mg/L	0.05	0.05	0.05	0.06	Non-pollution
Phosphate	0.5 mg/L	0.28	0.26	0.28	0.34	Non-pollution
Iron	0.3 mg/L	0.47	0.50	0.87	0.67	Slight pollution
Lead	0.01 mg/L	1.00	0.00	1.00	0.00	Slight pollution

Table 4.13: Water Quality Parameters and Pollution Grades across stations

Parameter	Regulatory Standard (Si)	Station 1 (Ci)	Station 1 Grade	Station 2 (Ci)	Station 2 Grade	Station 3 (Ci)	Station 3 Grade	Station 4 (Ci)	Station 4 Grade
pH	6.5–8.5	6.73	Slight pollution	6.73	Slight pollution	6.87	Slight pollution	6.87	Slight pollution
Electrical Conductivity (EC)	1000 µS/cm	83.00	Non-pollution	79.33	Non-pollution	81.33	Non-pollution	81.00	Non-pollution
Turbidity	5 NTU	8.32	Medium pollution	6.06	Medium pollution	8.49	Medium pollution	7.09	Medium pollution
DO	5 mg/L	6.53	Medium pollution	6.60	Medium pollution	6.33	Medium pollution	6.67	Medium pollution
BOD	6 mg/L	3.60	Slight pollution	3.47	Slight pollution	3.33	Slight pollution	3.53	Slight pollution
Nitrate	50 mg/L	2.67	Non-pollution	2.53	Non-pollution	2.68	Non-pollution	3.03	Non-pollution
Phosphate	0.5 mg/L	0.14	Non-pollution	0.13	Non-pollution	0.14	Non-pollution	0.17	Non-pollution
Iron	0.3 mg/L	0.14	Slight pollution	0.15	Slight pollution	0.26	Slight pollution	0.20	Slight pollution
Lead	0.01 mg/L	0.01	Slight pollution	0.00	Non-pollution	0.01	Slight pollution	0.00	Non-pollution

4.6 Chronic daily intake (CDI), Carcinogenic and Non-Carcinogenic Risk Assessment

Table 4.14 shows the chronic daily intake (CDI) of heavy metals via ingestion and dermal pathways. Iron (0.19 mg/L) and zinc (0.08 mg/L) had the highest concentrations, while arsenic, cadmium, and vanadium were below detection (0.00 mg/L). The ingestion route showed the highest CDI, with iron (5.99×10^{-3} mg/kg/day), zinc (2.52×10^{-3} mg/kg/day), and manganese (9.45×10^{-4} mg/kg/day) leading. Dermal absorption values were considerably lower, with zinc (2.96×10^{-6} mg/kg/day) and iron (9.37×10^{-7} mg/kg/day) being the most notable.

Table 4.15, presents the non-carcinogenic risks assessed through hazard quotients (HQs) and indicate that lead (2.25×10^{-1}) and chromium (2.23×10^{-1}) had the highest potential health risks. Other metals, including copper, iron, manganese, nickel, and zinc, had HQ values below 2.00×10^{-2}. Cadmium and vanadium posed no risk (HQ = 0.00). The total hazard index (HI) was 0.5148, below the acceptable threshold of 1.0, indicating no significant non-carcinogenic concern.

Table 4.16 presents the incremental lifetime cancer risk (ILCR) for adults. Mean ILCR values for chromium (2.58×10^{-2}), lead (2.68×10^{-3}), and nickel (2.65×10^{-4}) exceeded the acceptable limit of 1×10^{-4}. Maximum ILCR values for all carcinogenic metals also surpassed this threshold, while minimum values were zero. The cumulative cancer risk (ΣILCR) based on mean and maximum exposure levels were 2.87×10^{-2} and 5.95×10^{-2} respectively, both indicating an elevated long-term cancer risk.

Table 4.14: Chronic daily intake (CDI) for heavy metals through different pathways

Metal	Concentration (mg/L)	CDI (Ingestion) (mg/kg/day)	CDI (Dermal) (mg/kg/day)	CDI (Total) (mg/kg/day)
Arsenic	0.00	0.00	0.00	0.00
Cadmium	0.00	0.00	0.00	0.00
Chromium	0.02	6.30E-04	1.97E-07	6.30E-04
Copper	0.02	6.30E-04	9.86E-08	6.30E-04
Iron	0.19	5.99E-03	9.37E-07	5.99E-03
Manganese	0.03	9.45E-04	1.48E-07	9.45E-04
Nickel	0.01	3.15E-04	9.86E-10	3.15E-04
Lead	0.01	3.15E-04	4.93E-08	3.15E-04
Vanadium	0.00	0.00	0.00	0.00
Zinc	0.08	2.52E-03	2.96E-06	2.52E-03

Table 4.15: Non-carcinogenic human health risks posed by heavy metals in water of study area via different pathways.

Metal	Concentration (mg/L)	HQ (Ingestion)	HQ (Dermal)	HQ (Total)
Cadmium	0.00	0.00	0.00	0.00
Chromium	0.02	2.10E-01	1.31E-02	2.23E-01
Copper	0.02	1.58E-02	8.22E-06	1.58E-02
Iron	0.19	1.99E-02	3.12E-06	1.99E-02
Manganese	0.03	6.75E-03	1.06E-06	6.75E-03
Nickel	0.01	1.58E-02	1.83E-07	1.58E-02
Lead	0.01	2.25E-01	1.17E-04	2.25E-01
Vanadium	0.00	0.00	0.00	0.00
Zinc	0.08	8.40E-03	4.93E-05	8.45E-03
HI		0.5148		

Table 4.16: The incremental lifetime cancer risk (ILCR) values of carcinogenic human health risks via total exposure (ingestion and dermal contact) to the drinking water of the study area for adults.

Metal	Concentration (mg/L)	ILCR (Mean)	ILCR (Max)	ILCR (Min)	Implications
Lead (Pb)	0.01	2.68E-03	5.36E-03	0.00	Mean and Max: Unacceptable risk (ILCR > 1E-04). Min: Negligible risk.
Chromium (Cr)	0.02	2.58E-02	5.17E-02	0.00	Mean and Max: Unacceptable risk (ILCR > 1E-04). Min: Negligible risk.
Cadmium (Cd)	0.00	0.00	1.92E-03	0.00	Mean and Min: Negligible risk. Max: Unacceptable risk (ILCR > 1E-04).
Nickel (Ni)	0.01	2.65E-04	5.29E-04	0.00	Mean and Max: Unacceptable risk (ILCR > 1E-04). Min: Negligible risk.
Σ ILCR	—	2.87E-02	5.95E-02	0.00	Mean and Max: Unacceptable risk (ILCR > 1E-04). Min: Negligible risk.

CHAPTER FIVE

DISCUSSION

Microplastics have emerged as pervasive pollutants in aquatic ecosystems worldwide. In recent years, concerns over microplastics contamination have grown due to their persistence in the environment, potential for bioaccumulation, and associated risks to both ecosystem and human health. This study focuses on the assessment of microplastics in surface water and some fish species from the Ovia River, Benin City, Edo, Nigeria.

5.1 Physical and Chemical Parameters

The physical and chemical parameters recorded were compared with the WHO, USEPA, and FMEnv standards. Distinct spatial and temporal variations were recorded particularly in parameters sensitive to seasonal or anthropogenic influences. The results were consistent with findings from other studies across Nigeria and in the tropics (Ayanlade *et al*. 2019).

Water temperature varied subtly along the river continuum, with the highest temperatures observed upstream at Station 1, where the river flows through shaded zones dominated by oil palm (*Elaeis guineensis*) and rubber trees (*Hevea brasiliensis*). This canopy likely creates microclimates by limiting water mixing and trapping heat, resulting in slightly elevated temperatures compared to downstream stations. In contrast, downstream areas, characterized by increased flow and the presence of floating macrophytes likely contribute to marginal cooling effects. Seasonal variations showed the highest temperatures in March, coinciding with the dry-to-rainy transition period when solar radiation peaks before the cooling influence of the rainy season. Although these temperature variations fall within the acceptable range recommended by WHO and FMEnv guidelines, they can significantly influence the metabolic rates of aquatic organisms and the degradation rates of organic pollutants (Kumar *et al*., 2019; Agbaje *et al*., 2024). The pH measurements across the stations reflected a gradient from mildly acidic conditions upstream to near-neutral to slightly alkaline values downstream. Downstream stations (3 and 4), experience increased nutrient input from agricultural runoff and human activities, which promotes photosynthetic activity by floating macrophytes. This photosynthetic uptake of CO_2 raises the pH, shifting conditions towards neutrality or slight alkalinity. Temporally, the lowest pH values observed in March coincide with increased surface runoff during the dry-to-rainy seasonal transition, which dilutes the river's buffering capacity. Higher pH levels recorded in May align with peak aquatic plant growth during the rainy season. The pH values were near-neutral conditions and fall within WHO and FMEnv guidelines (6.5–8.5), and are consistent with tropical river studies (Val *et al.*, 2005; Jana, 2025), suggesting stable water chemistry that supports healthy aquatic ecosystems.

Electrical conductivity (EC) ranged from 81.00 to 83.00 µS/cm, indicating very low levels of dissolved ions and minimal contamination. Talling (2009) reported that EC values below 100 µS/cm are typical of relatively pristine freshwater systems, while Ntengwe (2006) observed that low conductivity is often associated with reduced pollutant loads. These studies support the finding that the Ovia River's EC levels are within the range expected for healthy aquatic environments. Total dissolved solids (TDS) measure the combined content of all inorganic and organic substances dissolved in water, serving as an indicator of overall water quality (Boyd, 2019). TDS values were significantly below the WHO limit of <500 mg/L (Kalevandi *et al*, 2024), suggesting minimal total dissolved solid contamination. Comparable studies in Nigerian water bodies (Zacchaeus *et al*., 2020; Olatunji *et al.*, 2021) have reported similarly low TDS levels, an indication that the water quality of the river is favorable regarding dissolved substances. The low TDS levels, particularly upstream (station 1 and 2) where anthropogenic disturbance is minimal, indicate a relatively unpolluted environment. Slight increases downstream (station 3 and 4) may reflect agricultural runoff and human settlements, yet values remain within acceptable ranges, affirming the overall favorable chemical status of the river

Turbidity is a measure of water clarity, reflecting the concentration of suspended particles that can affect light penetration and aquatic productivity (Boyd, 2019). Turbidity levels exceeded the recommended limit of <5 NTU (Chang and Liao, 2012). This elevated turbidity indicates higher suspended sediment loads or organic matter, which could be attributed to urban runoff, industrial discharges, or sediment resuspension. Comparatively, similar studies in Nigeria have reported elevated turbidity levels in urban and industrialized river systems. Alo *et al*. (2014) observed turbidity values exceeding 5 NTU in the Lagos Lagoon, attributing the high levels to urban runoff and improper waste management, while Ikotun *et al*. (2012) reported increased turbidity in the Ogun River due to industrial discharges and sediment disturbances. These findings reinforce that the Ovia River's turbidity levels is consistent with other urban Nigerian rivers and underscore the potential impact on aquatic ecosystems. Total suspended solids represent the concentration of particulate matter suspended in the water, affecting clarity, light penetration, and sediment deposition. In this study, TSS values were below the WHO limit of 30 mg/L suggesting that although turbidity was elevated, particulate pollution was relatively controlled. This could be attributed to sediment settling facilitated by the presence of vegetation like water lettuce and macrophytes downstream, which stabilize sediments and reduce suspended loads. Comparable research in Nigerian rivers (Titilawo *et al.*, 2018) has also reported low TSS levels, suggesting that sediment load is not a major factor affecting water quality in these systems.

Dissolved oxygen is a critical indicator of water quality, representing the amount of oxygen available for aquatic organisms (Best *et al*. 2007). DO levels exceeded the WHO minimum guideline of 5 mg/L (Kumar and Puri, 2012), and indicates favorable conditions for aquatic life. These levels likely result from the river's flow dynamics and photosynthetic activity by aquatic plants such as *Eichhornia crassipes* and *Nymphaea lotus*, which release oxygen into the water column. The maintenance of DO above critical thresholds despite increasing human activities downstream suggests resilience of the aquatic ecosystem. Comparable research in Nigerian urban water bodies has reported similar DO levels, supporting the observation that the Ovia River maintains adequate oxygenation despite anthropogenic pressures (Olatunde *et al,* 2015; Iyagbaye *et al*., 2017). Biochemical oxygen demand (BOD) reflects the amount of oxygen required by microorganisms to decompose organic matter in water. In this study, BOD values conformed with the WHO guideline of <5 mg/L indicating minimal organic pollution. Although slight spatial variations exist, the highest BOD values were observed at station 1 (upstream), likely due to organic debris and decomposing vegetation under the dense canopy of oil palm and rubber trees. Although station 1 experiences minimal direct human impact, the natural input of organic matter can elevate microbial activity. In contrast, station 3 (downstream) recorded the lowest BOD, possibly due to higher flow, sedimentation, and dilution of organic matter. Stations 2 and 4, both subject to human activities like fishing, farming, and washing, showed intermediate BOD values, suggesting a moderate but controlled organic load. The dry season in January had the lowest BOD, likely due to reduced organic input and minimal surface runoff. The transitional period in March brought increased temperature and organic flushing from initial rains, while the rainy season in May likely contributed to higher nutrient inflows and microbial decomposition. This observation aligns with Adekunle *et al*. (2017), who reported similar BOD trends in

Nigerian rivers affected by seasonal flow dynamics and organic inputs. Chemical Oxygen Demand (COD) measures the amount of oxygen required to oxidize both biodegradable and non-biodegradable organic matter in water. In this study, stations 2, 3, and 4 remained within the WHO permissible limit of <25 mg/L, while station 1 slightly exceeded this threshold, suggesting a relatively higher presence of oxidizable pollutants. COD increased steadily from January to March and peaked in May, with May values exceeding the WHO guideline. This trend reflects seasonal intensification of organic inputs runoff carrying domestic waste, decaying vegetation, and farming residues likely augmented oxygen demand, especially during the rainy season Similar studies in Nigeria, such as those by Kosemani *et al.* (2024) and Ojo (2024), have reported comparable COD levels in urban water bodies, linking them to domestic and industrial wastewater discharge.

Alkalinity, a measure of the water's buffering capacity against acidification, was low throughout the river, remaining below the WHO recommended range of 20–120 mg/L. Upstream stations (1 and 2) recorded the lowest values, likely due to limited carbonate input and acidic leachates from decomposing organic matter in the forested catchment. Downstream stations (3 and 4), though subject to anthropogenic runoff, did not show marked increases in alkalinity, suggesting insufficient input of bicarbonates from soils or domestic sources. Alkalinity rose gradually from January to March and reached the highest average in May. This progressive increase may reflect accumulating ions from surface runoff during the wet season, although values remained below optimal buffering levels. Low alkalinity during the dry season increases the system's vulnerability to acidification from organic acids or pollutant inputs. Similar low alkalinity levels have been observed in Nigerian rivers (Balogun *et al.*, 2024), emphasizing the need for ongoing monitoring to safeguard against potential acidification.

Salt-related parameters such as sodium, magnesium, potassium, and calcium are crucial for assessing water quality because they indicate the water's ionic balance and potential pollution sources. In this study, sodium, magnesium, potassium, and calcium across the four stations values conformed with the WHO guidelines (<200 mg/L for sodium and potassium, <30 mg/L for magnesium, and <75 mg/L for calcium). The upstream stations (1 and 2) showed slightly lower ion concentrations compared to downstream stations (3 and 4). Station 1, surrounded by natural vegetation and with minimal anthropogenic input, recorded the lowest values for most salts, indicating dominance of natural leaching from soil and bedrock. Station 2, though more impacted by domestic waste disposal and farming, still retained modest levels, suggesting minimal salt enrichment from those activities. Stations 3 and 4, located downstream and exposed to increased farming, fishing, bathing, and washing activities, showed marginal increases in sodium and calcium likely due to agricultural runoff and soil erosion but still remained within safe thresholds. This spatial pattern is consistent with the natural attenuation of ionic pollutants and the river's ability to buffer and dilute localized inputs. Salt concentrations were generally lowest in January (dry season), slightly increased in March (transition period), and peaked in May (rainy season). The dry season likely limited surface runoff and dilution processes, whereas the onset of rains in March and intensification in May likely mobilized ions from surrounding soils, agricultural fields, and settlements into the river system. However, even with this seasonal influx, concentrations remained consistently low, suggesting that salt inputs are not currently at levels that threaten aquatic health. These low and relatively consistent salt concentrations suggest that the Ovia River is not heavily influenced by salt pollution, in contrast to other urban rivers in Nigeria where industrial and domestic discharges often raise ionic levels (Udebuana *et al*, 2014;

Ayejoto *et al*, 2023). This stable ionic composition is critical for maintaining healthy aquatic ecosystems, as it supports the proper metabolic and physiological functions of aquatic organisms.

Water hardness, defined by the concentration of calcium (Ca^{2+}) and magnesium (Mg^{2+}) ions, is an essential parameter reflecting both geological influence and anthropogenic activities (Olajire and Imepeokparia, 2001). In this study, hardness levels across all stations remained well below the WHO recommended limit of 300 mg/l, indicating that the Ovia River exhibits very soft water characteristics throughout its course. Spatially, hardness values were lowest at Station 1 (upstream), located in a largely undisturbed environment with dense vegetation and minimal human interference. This suggests that geological formations in the area are not rich in calcium or magnesium-bearing minerals, and that anthropogenic input is minimal. Station 2, though influenced by domestic waste and farming, showed only slight increases in hardness possibly due to limited agrochemical use. Stations 3 and 4 (downstream) exhibited marginally higher hardness levels, potentially reflecting cumulative downstream effects from agricultural runoff and soil erosion, yet values still remained within soft water ranges. These downstream increments are not substantial enough to indicate significant pollution or geological leaching, reaffirming the natural softness of the river. Water hardness showed modest seasonal variations. The lowest values occurred in January (dry season), possibly due to reduced runoff and dilution from surrounding soils. March, marking the transition into the rainy season, showed a slight increase as early rains mobilized minerals from surface soils into the river. The highest hardness levels were observed in May (rainy season), coinciding with peak runoff and leaching from catchment soils, especially in the more anthropogenically active downstream stations. However, even during this period, total hardness remained significantly low, confirming that neither seasonal dynamics nor land use practices currently elevate hardness to concerning levels. Similar trends have been observed in other Nigerian water systems. Ogun Rivers have reported low hardness levels that were attributed to the regional geology and low industrial impact (Ojekunle *et al*., 2020). The consistency in our findings with those of other researchers shows that the Ovia River's water, like many Nigerian surface waters, exhibits naturally soft characteristics.

Chloride is a major anion that influences the taste and corrosiveness of water. It typically enters aquatic systems through both natural mineral weathering and anthropogenic inputs, such as agricultural runoff, industrial effluents, and in some regions, de-icing salts. In this study, chloride concentrations were consistently low across all stations and sampling months, remaining well below the WHO/USEPA threshold of <250 mg/L, indicating minimal chloride pollution. Spatially, concentrations were slightly higher at the upstream station and one midstream location, with lower values recorded at the remaining two stations. This suggests that certain locations may be more susceptible to influences from natural geologic sources or localized anthropogenic inputs such as domestic runoff and agricultural leachate. Temporally, chloride concentrations exhibited a clear increasing trend from the dry season (January) through the transition (March) to the rainy season (May). This rise is likely due to intensified surface runoff during rainfall, which can carry dissolved salts from the surrounding environment into the river system. Comparable chloride levels have been documented in urban and semi-rural water bodies in Nigeria. Studies by Popoola *et al*. (2019), Uzomah *et al*. (2021) and Ojo *et al*., (2025), reported similar low to moderate chloride concentrations, attributing these levels to natural sources and minimal industrial contamination Such comparisons support the view that the Ovia River exhibits chloride concentrations typical of Nigerian rivers not heavily impacted by industrial or urban pollution and is not subject to substantial chloride loading from human activities or saline intrusion.

The values recorded for nitrate concentrations suggest a uniform distribution of this parameter across the stations. These values are conformed with the WHO/USEPA guideline of <10 mg/l (Nangulu, 2015), indicating that nitrate contamination is minimal. Other research has shown that similar nitrate levels in rivers not heavily influenced by intensive agriculture or urban effluent. For example, studies on the lower Niger and certain tributaries have reported nitrate values within this safe range (Isiuku and Enyoh*,* 2020: Idriss *et al*., 2021). This is an indication that the Ovia River, like many other Nigerian surface waters, currently has limited nitrate enrichment. Sulphate is derived from the dissolution of minerals and can also result from anthropogenic sources. In this study, the measured sulphate levels values were exceedingly low when compared to the regulatory limits of <250 mg/l (WHO/USEPA) and even the stricter FMEnv guideline of <150 mg/l, indicating that sulphate is not a major concern in this water system. Spatially, all four stations exhibited similar nitrate values, indicating no significant point-source input. Temporally, nitrate levels showed little variation between the dry (January) and wet (May) seasons, a pattern that points to stable background levels rather than episodic or seasonal nitrate surges. Research on less impacted water bodies, such as certain streams in southeastern Nigeria, confirms that low sulphate levels are common when natural mineral dissolution predominates (Elueze *et al*., 2001). These comparisons further validate that the Ovia River's sulphate levels fall within the natural background range for the region. In contrast, phosphate concentrations were below the USEPA limit, they slightly exceed the more conservative WHO and FMEnv limit of <0.1 mg/l. This slight elevation may signal the onset of nutrient enrichment, which could have ecological implications if phosphate inputs continue or intensify. Alo *et al*. (2014) observed higher phosphate concentrations in the Lagos Lagoon, attributing the increases to improper waste management and runoff from urban areas. Similarly, studies on the Ogun River (Olasunbo *et al*,

2023) have reported phosphate levels that indicate the potential for eutrophication. The findings from the Ovia River align with these studies, suggesting that while the water remains within USEPA guidelines, there is a need for continuous monitoring to prevent long-term nutrient enrichment.

Heavy metals are an essential aspect of water quality assessments due to their potential toxicity even at low concentrations (Gheorghe *et al*. 2017). Notably, station 3 consistently exhibited the highest concentrations for several metals, suggesting it as a localized hotspot likely influenced by regional geology and anthropogenic inputs. Arsenic was below the detection limit (BDL) in all samples. Regulatory guidelines from WHO, USEPA, and FMEnv set the permissible limit at <0.01 mg/L. Cadmium levels were extremely low with the WHO limit at <0.003 mg/L (and USEPA at <0.005 mg/L), These results confirm that cadmium is not a major contaminant in this water. This aligns with findings from Iyasele and Idiata, (2012). Chromium concentrations levels had slight variation with higher values at some stations remains within the WHO guideline (<0.05 mg/l or, in some cases, <0.1 mg/l). Similar trends have been reported in studies of the Ikpoba and Osse Rivers, where chromium levels are generally low but may be slightly elevated at certain locations due to localized input (Igbinedion and Oguzie, 2016; Adams *et al*, 2016). Cadmium levels were extremely low well below the WHO limit of 0.003 mg/L. Station 3 recorded the highest copper levels compared with other stations. These values are well below the WHO limit (<0.2 mg/l) and are similar to reports from the Ovia and Ikpoba Rivers where copper is present at low concentrations (Igbinedion and Oguzie, 2016). Internationally, rural water bodies with limited industrial activity also show low copper levels, confirming that our observed values are within the expected safe range. Nickel was detected at very low levels, the observed nickel concentrations are well below the strictest guideline (<0.02 mg/l, with some standards allowing up to <0.1 mg/L), and similarly low levels have been observed in other Nigerian water studies (PJ and Don-Lawson, 2018; Shaibu and Audu, 2018). Nickel's low solubility in natural waters, combined with minimal anthropogenic input in our study area, contributes to these findings.

Among all metals, iron was the most abundant in the surface water samples. Although these levels approached the WHO limit of <0.3 mg/L, they still fall within acceptable bounds. Iron's predominance is typically due to its geogenic origin, as it readily dissolves from iron-rich minerals in the aquifer matrix. The dominance of iron over other heavy metals in aquatic ecosystem have been recognized by Puyate *et al*. (2007), Omoigberale *et al.* (2014) and Iwegbue *et al*. (2023). Manganese was observed between some stations, indicating localized influences perhaps due to differential dissolution from underlying rock types. However, all manganese levels remained within the WHO limit of 0.1 mg/L. Lead was present at trace levels; at two stations, while at others it was not detected and all values are at or below the WHO guideline of <0.01 mg/l. Given lead's high toxicity, these low levels are particularly encouraging. Previous studies in the Ikpoba Rivers have similarly reported very low lead levels, often attributing this to effective regulation and limited direct industrial discharges in certain areas (Iyasele and Idiata, 2012). Vanadium was below the detection limit (BDL) in all samples. With the guideline being <0.1 mg/l, this indicates that vanadium does not contribute to water quality concerns in our study area. Although zinc is essential in small amounts, excessive zinc can be harmful. However, our values are comfortably below the WHO guideline of <5 mg/l, this is similar with the findings of Shuaibu *et al*, (2015). The concentrations of most heavy metals (except manganese and copper) remained relatively stable across January, March, and May, with minor fluctuations possibly influenced by seasonal changes in runoff, soil erosion, and hydrological flow patterns. The slight increases in iron, manganese, and copper during the rainy season are consistent with mobilization of particles from surrounding soils and sediments.

The rank of heavy metals concentration in water was Iron > Zinc > Chromium > Copper >Manganese > Nickel > Lead, with arsenic and vanadium remaining below detection and cadmium effectively absent. In terms of inter-station variations, while most metals did not differ significantly among sampling points, copper and manganese showed slight statistical differences that could be attributed to localized factors such as differences in geology, runoff patterns, or minor point-source discharges. However, even the highest values recorded for these metals remained below their respective maximum permissible limits. This consistency suggests that, across the various stations, the water quality in terms of heavy metals is relatively uniform and is predominantly influenced by the underlying geology.

5.2 Microplastics

The characterization of microplastics across sampling stations and timeframes in the Ovia River revealed significant spatio-temporal variation in particle size distribution (PSD), abundance, and likely sources. This variation reflects the dynamic nature of plastic pollution in freshwater ecosystems and the role of both natural processes and anthropogenic inputs.

During the dry season, when water flow was relatively low and dilution effects minimal, stations 1 and 4, characterized by minimal disturbance and partially intact riparian vegetation, displayed a tendency toward more uniform microplastic particle sizes. These uniform particles patterns are indicative of localized accumulation with limited external inputs and slower degradation processes. The stability of the aquatic environment at these stations likely allowed for the persistence of older microplastic particles with less mechanical fragmentation, reflecting background pollution levels from low-impact sources such as organic debris and minimal runoff. In contrast, station 2, which receives direct domestic waste discharge and supports activities like fishing and farming, presented evidence of a more variable particle size distribution even during the dry season. This suggests continued input of fresh plastic debris, likely from household materials and discarded packaging, which may not have undergone extensive environmental degradation. Similarly, station 3, surrounded by active farmland and subject to multiple land-based activities, showed signs of microplastic input from agricultural sources, though the relatively still water of the dry period may have temporarily limited further fragmentation.

As the system entered the transitional season, marked by intermittent rainfall and the start of land preparation and planting activities, increased runoff introduced greater heterogeneity in particle sizes especially at stations 2 and 3. These stations, already under substantial anthropogenic influence, experienced a flush of both recent and aged plastic fragments, possibly carried in from farmlands, homesteads, and nearby disturbed banks. The interaction of fresh inputs with previously settled plastics likely created a broader range of particle sizes, reflecting both newly introduced polymers and partially degraded ones mobilized from sediments.

During the rainy season, increased turbulence, surface runoff, and sediment resuspension contributed to the overall reduction in particle sizes across the system. This was most evident at stations 2 and 4, which are more accessible and exposed to intense human use such as bathing, washing, and waste dumping. The resuspension of buried microplastics and the continued influx of fragmented waste materials along with enhanced physical abrasion likely accelerated the breakdown of larger plastic debris into smaller fragments, resulting in a wider dispersion of fine particles. Even at station 1, the rainy season introduced variability likely due to overland flow from upstream disturbances, despite its relative isolation from direct human activity. Station 3, though primarily influenced by agriculture, also experienced increased particle size diversity during the rainy season, likely due to agro-runoff containing degraded films, sacks, and packaging materials. These findings underscore the significance of land use and rainfall in modulating both the physical characteristics and transport of microplastics in aquatic environments. Across all stations and seasons, the rejection of some PSD data based on the relationship between mean values and standard deviations highlights instances of extreme variability, particularly during periods of high runoff or sediment disruption. This irregularity points to episodic pollution events and the presence of microplastics at various stages of environmental degradation, from relatively intact materials to highly fragmented, aged particles. This trend, which aligns with findings from Browne *et al*. (2010); Dris *et al*. (2015), and Adeogun *et al*. (2020), indicates that prolonged exposure to UV radiation, turbulence, and microbial degradation leads to continuous fragmentation, resulting in an abundance of finer particles.

In summary, station 1 generally reflected minimal impact, station 2 showed persistent and direct anthropogenic influence, station 3 mirrored the effects of intensive agricultural activity, and station 4 revealed dynamic interactions between community usage and natural sediment processes. These spatial patterns, along with seasonal hydrology, reveal the complex interplay between human behavior, environmental forces, and plastic degradation pathways. The findings underscore the need for context-specific interventions such as waste management near populated zones and erosion control in agricultural areas to curb microplastics contamination in freshwater ecosystems.

The SEM analysis of fish samples reveals that both *Notopterus afer* and *Heterobranchus bidorsalis* exhibit a clear temporal trend of decreasing mean particle size. During the dry season, when sediment disturbance is minimal and water clarity is higher, both species ingested microplastics that tended to be relatively larger and more uniform in size. Furthermore, larger microplastic particles may have settled more prominently on benthic substrates or become entangled in vegetation and detritus, making them more available to bottom-dwelling species like *Heterobranchus bidorsalis*, a benthic omnivore with scavenging tendencies. Similarly, *Notopterus afer*, which also forages near the substrate, may have encountered and ingested microplastics during bottom feeding or detrital consumption. This pattern aligns with findings from Wu *et al.* (2022) in Lake Chaohu, China, where higher microplastic concentrations were observed in fish during the dry season, attributed to limited dilution and increased accumulation of particles.

In the transitional season, the particle sizes retrieved from both species became more variable. This period typically marks the beginning of rainfall, leading to intermittent sediment agitation and increased runoff. These hydrological changes likely introduced a mixture of newly fragmented microplastics and previously settled particles into the water column. The presence of particles at different stages of degradation within the feeding grounds of both species may have resulted in a broader size range within their gastrointestinal contents. Similar seasonal variability in microplastic characteristics has been documented by Onay *et al*. (2023) in fish from the Southeast Black Sea, where differing polymer types and shapes were observed across seasons.

During the rainy season, the prevalence of smaller microplastic particles in both fish species became more apparent. Higher flow rates and persistent sediment resuspension during this period tend to break down larger plastic debris into finer fragments. Additionally, increased runoff from surrounding land carrying degraded plastic waste contributes to the influx of small-sized particles into the aquatic system. The higher ingestion of these finer particles may not only be a function of environmental availability but also the feeding behavior of the fish. *Heterobranchus bidorsalis*, due to its bottom-feeding and scavenging nature, likely encountered these resuspended microplastics within disturbed sediments. In contrast, *Notopterus afer* may have consumed smaller particles incidentally while feeding on invertebrates or detritus within mid to lower water columns. The reduced particle size range in both species during this season suggests intensified degradation and dispersal mechanisms acting on microplastics within the aquatic environment. This observation is consistent with the findings of Zhang *et al*. (2021), who reported that fish in regions with higher hydrodynamic activity tend to ingest smaller microplastic particles due to increased fragmentation and dispersion. Despite the differences in foraging habits and ecological niches, both species exhibited similar seasonal trends in particle size distribution. This suggests that environmental conditions and particle availability, rather than species-specific selectivity, primarily govern the size of ingested microplastics in this system. Supporting this, Lopes *et al*. (2020) demonstrated that microplastic ingestion in fish is more influenced by environmental exposure and availability than by feeding strategies alone.

Overall, the dynamic spatial and temporal patterns observed in the PSD of microplastics underscore the complex interplay between pollution sources and environmental degradation processes in the Ovia River. These results reinforce previous studies (Andrady, 2011; Cole *et al*., 2011) and highlight the need for targeted management strategies to address microplastics pollution in freshwater ecosystems.

Polymeric Composition of Microplastics

According to Bayo *et al.* (2021) there are two regions of FTIR spectroscopy spectra that are the fingerprint region (between 600 and 1500 cm^{-1}) and the functional group region. However, the functional group is composed of single bond region (2500–4000 cm^{-1}), triple bonds (2,000–2,500 cm^{-1}), and double bond region (1,500–2,000 cm^{-1}) wavelength. The fingerprint region is complex and is composed of a large number of peaks, therefore, it was not used as opposed to the functional group region (Smith, 2011). In this study the functional group regions were between 1,500 and 4000 cm^{-1}. Characteristic absorption bands such as O–H stretching (3300–3400 cm^{-1}), C=O stretching (1630–1640 cm^{-1}), and CH_2/CH_3 stretching (2850–2920 cm^{-1}) were indicative of common polymers. These distinct differences were observed across the study stations. Across all the study stations and months, the recurring detection of polyvinyl alcohol (PVA) was prominent, particularly characterized by strong O–H stretching peaks. This polymer, known for its water solubility and frequent application in packaging films, adhesives, and textile sizing, appeared consistently in the dry season (January), transitional period (March), and rainy season (May), suggesting a persistent input source. Its ubiquity points to a combination of domestic wastewater and industrial effluents as likely contributors, particularly since PVA is extensively used in laundry pods, paper coatings, and personal care products (Chiellini *et al.*, 2003). Its dominance across all stations further reflects its widespread use and resistance to degradation in aquatic environments.

The identification of polyamide (nylon) through characteristic C=O stretching vibrations across all months and study stations further underscores the widespread use of synthetic fibers, likely entering the aquatic environment through washing of synthetic textiles or fishing gear degradation. Given its robust mechanical properties, nylon is frequently used in fishing nets, ropes, and synthetic fabrics, and its constant presence across the seasons implies both local and continuous sources, including artisanal fishing and domestic discharges. Nylon, like PVA, consistently emerged as one of the most frequently occurring polymers, further solidifying its role as a key indicator of anthropogenic textile influence in surface waters which is consistent with studies such as Browne *et al*. (2010) that documented high levels of synthetic fibers in polluted aquatic systems.

Polyethylene variants were also prevalent, particularly low-density polyethylene (LDPE), high-density polyethylene (HDPE), and polyethylene glycol (PEG), all indicated by CH_2 and CH_3 stretching and O–H stretching for PEG. These materials, widely used in plastic bags, bottles, medical products, and packaging materials, appeared predominantly in March and May, suggesting enhanced transport into the water bodies during rainfall and surface runoff. Stations 2 and 3, which receive significant input from agricultural lands and household discharges, exhibited the most frequent detections of these polyethylene types. The seasonal influx of such polymers underscores the hydrological role in mobilizing surface litter and medical-related plastics into the aquatic system. Even in station 1, where such inputs would be minimal, trace amounts were observed, suggesting downstream transport or aerial deposition. While not as dominant as PVA and nylon, their steady presence across multiple stations highlights the role of seasonal surface wash-off in mobilizing packaging and medical-related polymers. Their distribution further implies a direct linkage to urban and peri-urban waste pathways, especially during periods of high rainfall and catchment runoff (Li *et al*., 2018).

In addition, polypropylene (PP), also identified through CH_2 and CH_3 stretching, emerged more prominently during the transitional season in station 1 and 2. This polymer, often derived from bottle caps, disposable containers, and fabric blends, exhibited moderate abundance across several sites. Its buoyant nature and widespread utility make it a recurring but less dominant microplastic type, indicative of generalized domestic waste contributions. More notably, the detection of polyurethane and polyethylene terephthalate (PET) in January and March in station 2 and 3, suggests contributions from more structurally complex materials such as foam padding, construction coatings, and beverage bottles. These materials typically originate from consumer waste or construction debris, and their presence during drier months could reflect accumulation from low water flow conditions, which concentrate surface contaminants in localized areas, especially at stations exposed to domestic waste or minimal flow turnover. Though less frequent, their detection signals the infiltration of more durable polymer types into the aquatic system.

Unique polymers such as polyacetylene and cellulose (cellophane) were identified primarily at stations surrounded by farmlands and dense macrophytes (station 3 and 4). Polyacetylene, with C≡C stretching suggests less common industrial or agricultural plastic sources. Its detection may point to specialized films or agricultural mulch. Meanwhile, Cellophane and PEG, both hydrophilic and biodegradable to some extent, likely originate from food packaging and pharmaceutical industries. Their identification, especially in May in station 3, could also signal a surge in biodegradable plastic use or runoff from urban and semi-urban areas during peak rainfall. While these polymers were not among the most abundant, their presence is ecologically important due to their potential to degrade differently and interact with aquatic organisms in unique ways (Acharya *et al*. 2021).

Seasonal trends further emphasize how environmental conditions mediate polymer presence. During the dry season (January), polymer types such as nylon, PVA, and polyurethane dominated, implying limited water movement and localized accumulation of anthropogenic inputs, particularly from fishing gear, domestic residues, and degraded packaging. As the environment transitioned in March, the appearance of polypropylene, LDPE, and PET marked a shift toward greater diversity and mobility, possibly enhanced by early runoff events and agricultural activity beginning in surrounding lands. The rainy season (May) heightened the transport and distribution of polymers such as PEG, cellulose (cellophane), and HDPE, indicating an increase in surface wash-off, flooding-induced sediment disturbance, and higher pollutant influx into the aquatic ecosystem. This aligns with the findings of Dris *et al*. (2015), who observed a wide range of polymer types in urban waterways

The presence of these polymers, particularly those known for hydrophobic surfaces such as LDPE and PP, is ecologically significant because of their tendency to adsorb and transport chemical pollutants and heavy metals (Agboola and Benson, 2021). Their coexistence with trace elements, as observed in the elemental analysis, points to a synergistic pollution mechanism where microplastics act not only as physical contaminants but also as vectors for chemical substances, including nutrients, metals, and potentially toxic compounds. This capacity to concentrate and carry adsorbed substances further elevates their ecological risk, particularly during the rainy season when nutrient loading, pesticide runoff, and wastewater inputs peak.

In sum, the polymeric diversity observed across stations and seasons reflects a complex interplay of anthropogenic activities, environmental conditions, and hydrological dynamics. The dominance of PVA and nylon across all study stations indicates a consistent source of synthetic fibers and water-soluble plastics (Kedzierski *et al*. 2020), while the seasonal appearance of polymers such as PET, PEG, and Polyurethane highlights the role of human activities and climatic events in modulating microplastic types in surface waters. These findings corroborate broader global studies (Cole *et al*., 2011; Du *et al*., 2021; Zhang *et al*., 2021), emphasizing that even freshwater systems in less industrialized regions are susceptible to a wide spectrum of plastic pollutants with significant ecological implications. In Nigeria, research by Adeogun *et al.* (2020) study on Lagos lagoon and Idowu *et al*. (2024), has similarly identified high levels of nylon and polyethylene, attributing these findings to extensive use of sachet water packaging and textiles. Li *et al*, (2018) also reported the similar findings but PVC were also dominant in water samples.

In fish (*Notopterus afer* and *Heterobranchus bidorsalis),* a total of five polymers were identified. Polypropylene (PP) and Low-Density Polyethylene (LDPE) were the most consistently identified polymers in both species, underscoring the ubiquity of packaging-related plastic debris in the aquatic environment This composition reflects a consistent background input of urban and domestic waste, as noted in studies by Browne *et al*. (2010). Their spectral signatures marked by characteristic CH_2 and CH_3 stretching vibrations appeared uniformly in January, March, and May, indicating a persistent exposure of both fish species to non-biodegradable plastic materials likely originating from consumer waste and domestic sources.

Notopterus afer exhibited a consistent presence of Polyethylene Terephthalate (PET) in the dry and transitional months. As a commonly used polymer in beverage containers and food packaging, its identification may reflect direct ingestion of fragments or secondary microplastics resulting from degradation within the water column or sediment. Its reduced detection by May aligns with increased water volume and flow during the rainy season, which may dilute or disperse PET particles or shift the exposure pathways for fish.

A shift in polymer profile was observed in May with the emergence of Polyvinyl Alcohol (PVA) in *Notopterus afer*, suggesting seasonal variability in polymer availability or uptake. This water-soluble polymer, frequently associated with packaging films and laundry-related products, appeared during the rainy season, likely facilitated by surface runoff and increased inflow from domestic effluents. The presence of PVA in fish tissues during this period highlights the role of seasonal hydrodynamics in modulating microplastic exposure, particularly for soluble or low-density polymers. This is similar to Jabeen *et al,* (2017) work on various fish freshwater *spps.*

In *Heterobranchus bidorsalis*, while PP and LDPE remained dominant throughout, additional polymers such as cellulose (cellophane) and PVA emerged in the transitional and rainy seasons. The detection of cellophane, a regenerated cellulose material used in food packaging during March is particularly noteworthy. Its presence, alongside PVA, underscores a broader range of polymer exposure in this species compared to *Notopterus afer*, possibly due to differences in feeding behavior, habitat utilization, or sediment interaction. Given that *Heterobranchus* is an omnivorous, benthic species that often scavenges at the bottom, its contact with settled or buried microplastics may result in the ingestion of a more diverse polymer range. This is consistent with the findings of Park *et al.* (2022) and da Costa *et al*. (2023).

Seasonally, January's polymer profile in both species was dominated by conventional, hydrophobic plastic types such as PP, LDPE, and PET, consistent with reduced water movement and sediment resuspension. In contrast, March and May marked a shift toward hydrophilic and water-soluble polymers, suggesting that rainfall and surface inflow facilitated the mobilization and ingestion of more varied plastic types, particularly those derived from household and packaging waste. The co-occurrence of these polymer types across both fish species points to shared exposure pathways but also highlights species-specific differences in microplastic assimilation and possibly trophic behavior. Xu *et al.* (2019) documented that seasonal variations such as changes in rainfall and river flow can alter the composition of pollutants entering the water, while differences in feeding behavior and digestive processes between the two species may influence the retention and breakdown of microplastics (Sun *et al,* 2019). The dominant polymers PP and LDPE, found in this study are consistent with those reported in Ikopba River by Ogbomida *et al*. (2023). Ghosh *et al*. (2021) and Pozo *et al*. (2019) also recorded that PP, PE, PET and polyamide were identified as the prevalent polymers detected. Research conducted by Rochman *et al*. (2013) established that microplastics, primarily comprising LDPE and polypropylene, have become ubiquitous in aquatic ecosystems due to improper waste management practices and urban runoff.

Taken together, the temporal and interspecies polymer profiles suggest that aquatic biota in the Ovia River system is exposed to a persistent and seasonally modulated spectrum of microplastics, reflecting the complex interplay between plastic pollution sources, hydrological conditions, and ecological habits of resident fauna. These findings not only demonstrate the pervasive nature of plastic contamination in freshwater fish but also offer insight into the environmental behavior of specific polymer types and their ecological reach.

Elemental Composition of Microplastics

The EDX analysis provides insight into the elemental composition of microplastics collected from surface water and fish samples at different stations and sampling times. The elements detected help in understanding possible contamination sources and interactions between microplastics and environmental pollutants. According to Campanale *et al*. (2020). The vast surface area of microplastics makes it an ideal medium for the adsorption of hazardous compounds such as heavy metals and persistent organic pollutants. The EDX analysis of microplastics in surface water revealed a complex elemental profile that included chlorine (Cl), nitrogen (N), sodium (Na), carbon (C), calcium (Ca), magnesium (Mg), silicon (Si), phosphorus

(P), potassium (K), aluminum (Al), iron (Fe), sulfur (S), titanium (Ti), silver (Ag), and manganese (Mn).

In the dry season (January), elemental profiles at less disturbed sites like station 1 showed moderate levels of carbon, nitrogen, chlorine, and sodium, suggesting a background presence of organic and inorganic matter bound to microplastics. The relatively low water flow during this period allows accumulation and stronger adsorption of elements onto micro plastic surfaces. Conversely, stations with increased human activity (Stations 2 and 3) recorded a broader suite of elements, including iron, aluminum, silicon, and sulfur, which can be attributed to agricultural runoff, domestic waste, and soil erosion. These elements adsorbed onto microplastics serve as indicators of localized pollution and may facilitate the transport of pollutants downstream. The sparse vegetation and presence of decaying organic materials further contribute to the complexity of elemental interactions in this area. At station 4, the elevated presence of carbon suggests an abundance of organic matter in the water, likely derived from the continuous decay of plant material, as the station's substrate is composed of decomposing vegetation mixed with clay. This organic-rich environment creates favorable conditions for the colonization and accumulation of microplastics, which often interact with natural organic matter through surface adsorption. The detection of elements such as chlorine and nitrogen during this season may be attributed to household activities like washing and bathing, which, though seemingly low-impact, introduce detergents, soaps, and residues directly into the aquatic system. These chemicals often contain chlorine-based compounds and nitrogenous materials, including nitrates and ammonia derivatives, which readily adhere to microplastic surfaces. The relatively undisturbed flow of water in the dry season may allow microplastics and their associated elements to remain suspended in the water column for extended periods, enhancing opportunities for sorption of dissolved elements. As the system transitioned from dry to rainy season in March, elemental dominance shifted markedly. Both station 1 and station 3 displayed a striking increase in carbon content, underscoring the influence of organic matter dynamics in relatively vegetated zones with low to moderate human disturbance. At station 1, where oil palm and rubber trees provide dense canopy cover and aquatic plants like *Nymphaea lotus* thrive, the sharp dominance of carbon is indicative of accumulated organic inputs, likely from leaf litter, decomposing vegetation, and stable biofilm layers on microplastic surfaces. Similarly, at station 3 characterized by rich muddy substrates and extensive aquatic macrophytes such as *Salvinia sp. Nymphaea sp. Lemna sp.* and *Eichhornia crassipes*, the elevated carbon levels reflect a synergistic process of organic decay and microbial colonization. Biofilms, which are microbial communities embedded in extracellular polymeric substances, often colonize microplastics in aquatic environments, contributing additional carbonaceous material (Zettler *et al*., 2013). A similar trend was observed at station 4, where carbon content also peaked during March, corresponding with the presence of submerged vegetation and decomposing plant matter in the substrate. These conditions create an ideal setting for biofilm development and microbial activity, reinforcing the role of microplastics as vectors for transporting carbon-rich organic pollutants and microorganisms within the aquatic system.

At station 2, the elemental profile for March deviated significantly from the other stations. This site, characterized by high levels of anthropogenic disturbance such as farming and domestic waste disposal, showed an unusual spike in sulfur, titanium, and silver. The presence of these elements suggests early runoff events mobilizing materials from fertilizers, wastewater, or even electronic waste, which then enter the waterway. Microplastics, due to their hydrophobic surfaces and capacity to adsorb heavy metals and other contaminants (Yu *et al*, 2019; Naqash *et* *al*. 2020), may play a pivotal role in sequestering and transporting these pollutants. The elevated silver, in particular, could indicate contamination from antimicrobial agents or industrial discharge, a trend that has been documented in rivers near human settlements (Nowack, 2010). These findings underscore the dual threat of microplastics not only as persistent particles in the environment but also as vectors for trace metals and harmful compounds, especially during the early rainy season when runoff inputs are most pronounced. Furthermore, titanium, though present in small amounts, may originate from urban sources such as sunscreen, paints, or plastic additives, which bind readily to microplastic particles

During the peak of the rainy season in May, increased rainfall and runoff substantially altered the elemental composition across the stations. At Station 1, the composition reflected a marked rise in chlorine and sodium, alongside detectable levels of silicon, phosphorus, and aluminum. These elements likely originated from surface wash-off of decaying vegetation, organic debris, and localized natural leaching from the sandy substratum. The presence of aluminum and phosphorus may be associated with the breakdown of leaf litter and organic-rich particles, while silicon could be sourced from soil erosion and suspended sediment input. Microplastics in this setting likely served as binding platforms for these materials, aided by enhanced biofilm formation due to nutrient enrichment and microbial activity. At station 2, the elemental profile was more complex, with notable increases in sodium, silicon, phosphorus, potassium, and aluminum, accompanied by trace amounts of calcium, magnesium, and iron. This station, characterized by farming and domestic waste discharge, presented signs of nutrient loading and pollutant influx. The surge in silicon and aluminum suggests considerable terrestrial input, likely from soil erosion and fertilizer runoff from nearby farms. The presence of phosphorus and potassium points to agrochemical use, while elevated sodium is consistent with detergent-laden effluents from washing activities. The association of these elements with microplastic surfaces underlines their role as adsorptive carriers in a highly disturbed zone.

At station 3, the rainy season brought about elevated chlorine and sodium concentrations, alongside silicon, phosphorus, aluminum, and iron. These elements appear to be mobilized from surrounding farmlands and organic-rich sediment. The presence of phosphorus and iron in measurable quantities indicates a possible contribution from decaying biomass and redox-driven mobilization from sediments under increased hydrological disturbance. These findings agree with Bhat and Janaszek (2024), who reported similar elemental profiles in the Nida River. Microplastics in this context likely acquired these elements through interactions with fine particulates and dissolved contaminants, further amplifying their ecological threat.

Station 4, which experiences regular fishing, washing, and bathing activities, exhibited high levels of sodium and chlorine during this period, with supporting contributions from silicon, calcium, aluminum, and phosphorus. The deforested riverbank and clay-rich substrate likely facilitated the mobilization of aluminum and silicon, while calcium and phosphorus may derive from detergent use and decomposing plant matter. Gopinath *et al*, (2020) also found various metal present such as Al, Fe, Si, Ti which was similar to our findings. The interplay between anthropogenic input and natural runoff resulted in microplastics becoming complex vectors of multi-sourced elemental contamination.

Across all stations, the rainy season intensified the interaction between land-based activities and aquatic chemistry, with microplastics acting as dynamic participants in the transport and redistribution of elements. Unlike the drier months, where carbon or isolated elements dominated, the rainy season encouraged a more diverse elemental load onto microplastic particles. These particles, influenced by surface charge, roughness, and biofilm development, readily attracted a broad spectrum of metals and nonmetals, transforming them into potential sources of both chemical stress and ecological disturbance. The absence of silver during this period suggests a dilution or non-persistence of some trace contaminants, but the elevated macro- and micronutrient levels pose their own environmental challenges, particularly for aquatic life sensitive to nutrient imbalance or metal toxicity.

For fish samples, the presence of high calcium and phosphorus levels in both species, particularly evident in May during the rainy season, strongly indicates biological accumulation related to the fish's skeletal structures, such as bones and scales, or dietary uptake. These elements are natural constituents of fish physiology, reflecting the biomineralization process that varies somewhat between species due to differences in habitat and feeding behavior.

Heterobranchus bidorsalis, a benthic feeder, interacts closely with sediment environments where microplastics tend to accumulate alongside mineral-rich particles. This benthic foraging behavior facilitates direct exposure to microplastics carrying adsorbed elements like iron, aluminum, and silicon, as well as calcium and phosphorus. The benthic zone's organic-rich, fine silt substrate, particularly in stations surrounded by farmlands, likely enhances the retention and transfer of these elements to microplastics, which are subsequently ingested by *Heterobranchus* during bottom feeding. This interaction results in the relatively higher calcium and trace metals observed in this species, highlighting the role of sediment-associated microplastics in contaminant bioaccumulation. The increase in phosphorus alongside calcium in May corresponds with the rainy season's intensified runoff, which mobilizes both natural and anthropogenic materials, including agricultural fertilizers rich in phosphorus compounds. These runoff events elevate the elemental load available to microplastics, intensifying the risk of pollutant transfer to benthic feeders.

Notopterus afer, generally a midwater or surface feeder, is less directly associated with sediment but can ingest microplastics through the water column or trophic transfer via its prey. Although it feeds away from the sediment, the elemental profiles show similar calcium and phosphorus trends, implying ingestion of biologically relevant materials alongside microplastics. The presence of silicon and iron in both fish species suggests ongoing exposure to microplastic-contaminated water, likely influenced by suspended particulate matter and sediments resuspended during periods of increased flow and rainfall. This is consistent with findings from other studies (Du *et al*. 2021) where microplastics act as vectors for various elements including metals and nutrients, adsorbed from contaminated waters and sediments. These findings underscore the complexity of microplastic contamination pathways in freshwater systems and the influence of species-specific feeding strategies.

Seasonal variation further shapes these elemental patterns. During the dry season (January), lower water flow reduces sediment disturbance and runoff, limiting the availability of sediment-bound elements on microplastics. This period corresponds to generally lower elemental concentrations on microplastics in both fish species, reflecting more stable environmental conditions and reduced pollutant mobilization. However, as the system transitions from dry to rainy season in March, increased runoff begins to introduce higher levels of suspended solids and associated elements, which adhere to microplastics, increasing their contaminant load. By May, the peak rainy season, intensified runoff and sediment mobilization result in complex elemental patterns on microplastics, including elevated calcium, phosphorus, silicon, aluminum, and trace metals. These seasonal fluxes illustrate the dynamic nature of microplastic contamination and its potential to vary with hydrological and ecological conditions.

Furthermore, the elemental composition in surface waters surrounding these fish reinforces the interpretation of biological and environmental sources of contamination. High sodium and chlorine levels in water samples indicate persistent domestic or industrial pollution, while silicon and aluminum suggest sediment resuspension and possibly anthropogenic contributions related to land use, such as agriculture and construction activities (Zhang *et al*. 2025). The concurrence of phosphorus in both surface water and fish microplastic samples likely reflects fertilizer runoff and wastewater inputs, further supporting the interconnectedness of environmental and biological contamination pathways.

Together, these results demonstrate that microplastics in freshwater fish are not merely passive contaminants but active vectors that accumulate diverse elemental loads depending on environmental context and species-specific ecology. The interplay of hydrological seasonality, land use, and feeding behaviors shapes the contamination profiles, highlighting the multifaceted risks microplastics pose to aquatic organisms. This complex interaction suggests that mitigation efforts must consider not only microplastics presence but also their role in transporting chemical contaminants across ecological boundaries.

5.3 Water Quality Index (WQI)

The Water Quality Index (WQI), derived from a suite of physicochemical parameters such as temperature, pH, electrical conductivity, turbidity, total suspended solids, total dissolved solids, dissolved oxygen, biochemical oxygen demand, chemical oxygen demand, alkalinity, nutrients (nitrate and phosphate), and trace metals, offers an integrated assessment of river health. Spatial differences in water quality were evident across the study stations. Upstream areas, particularly the minimally disturbed station 1, generally exhibited better water quality conditions, suggesting the presence of effective self-purification processes, relatively intact riparian vegetation, and limited human interference. These factors collectively contribute to the dilution and assimilation of pollutants. In contrast, stations located downstream, especially stations 2, 3, and 4, showed progressively poorer water quality, strongly influenced by human activities such as farming, domestic waste disposal, open defecation, fishing, and washing. These practices introduce nutrients, organic matter, and potentially heavy metals into the river system, altering its ecological balance. Station 3, which is surrounded by intensive agricultural activity and has a silty, organic-rich substrate, appears particularly vulnerable to contamination. Its location within a farming-dominated landscape with reduced vegetation cover may facilitate runoff laden with fertilizers, organic waste, and eroded soil particles. The accumulation of suspended solids and increased nutrient loading at this site likely promotes the formation and retention of microplastics, which often bind with fine sediments and organic matter. A similar seasonal trend has been observed in other parts of Nigeria, where wet season runoff leads to the enrichment of water bodies with nutrients and organic pollutants, thereby lowering water quality. Ikhile and Aderogba (2011) reported that rivers in southwestern Nigeria experienced higher biochemical oxygen demand and lower dissolved oxygen during the rainy season, consistent with elevated pollution levels.

The presence of trace metals, though within acceptable limits, underscores potential long-term ecological risks. Even at low concentrations, elements like iron, copper, and manganese may adsorb onto microplastic surfaces, increasing their chemical reactivity and ecological threat (Naqash *et al*. 2020). Such interactions can lead to the formation of contaminant mixtures, where microplastics serve as carriers for heavy metals, enhancing their persistence and bioavailability in aquatic systems. The deteriorating conditions downstream suggest cumulative pollution effects and raise concerns about the sustainability of aquatic ecosystems and water use for local communities. Furthermore, water quality parameters such as turbidity, nutrient levels, and organic content influence the behavior of microplastics, including their transport, aggregation, and interaction with biota. For instance, high turbidity and organic matter can enhance microplastic clustering, while low dissolved oxygen conditions may increase the vulnerability of aquatic organisms to microplastic ingestion and toxicity. Therefore, continuous monitoring of water quality, particularly during the rainy season and in areas with intense human activity, is crucial.

5.4 Comprehensive Pollution Index

The Comprehensive Pollution Index (CPI) provides an integrative evaluation of water quality by combining several parameters, including pH, electrical conductivity, turbidity, dissolved oxygen, biochemical oxygen demand, chemical oxygen demand, alkalinity, nutrients such as nitrate and phosphate, and trace metals like iron and lead. This index captures both the presence and intensity of pollutants in the aquatic system and is particularly useful for detecting cumulative or low-level pollution pressures. In the present study, the CPI results indicated that all sampling stations within the study stations fell into the slightly polluted category, signifying persistent but moderate contamination across the river system. This classification reflects the influence of diffuse pollution sources and a mixture of both natural and anthropogenic inputs, consistent with observations in other Nigerian rivers such as the Ogun and Osun, where similar CPI classifications have been reported (Adefemi *et al*. 2007; Agbebaku, 2021). Spatial patterns in pollution levels reveal that downstream stations, especially those influenced by human settlements and agricultural activity, show greater cumulative impacts. Station 1, located in an upstream region with minimal disturbance and dense vegetation, and exhibited relatively better water quality conditions. The CPI result for this station suggests limited pollutant input, likely due to effective natural filtration and low levels of runoff. Similar findings have been documented by Adamu *et al*. (2016), who noted that CPI scores tend to be lowest in headwater regions with fewer anthropogenic activities. Station 2, situated in a moderately disturbed area with domestic waste disposal and some farming activities, showed a more complex pollution profile. Though not the most degraded, this station reflects signs of increasing stress likely associated with localized sources such as washing, fishing, and unregulated waste inputs. Station 3, which lies in the heart of agricultural activity and is surrounded by farmlands cultivating crops like cassava, yam, and maize, was the most impacted in terms of CPI. The silty substrate and low vegetative cover promote surface runoff rich in agrochemicals and organic matter, contributing significantly to water quality deterioration. Station 4, located downstream and influenced by human activities such as washing, bathing, and deforestation, showed moderate pollution levels. Though not as severely impacted as station 3, the loss of forest cover and organic debris loading may contribute to chemical and particulate inputs. This scenario reflects trends seen in other tropical freshwater systems where downstream areas accumulate pollutants from upstream and adjacent sources (Sharma and Kansal, 2011). The CPI findings correlate well with the Water Quality Index assessments, confirming that areas with higher pollution burdens also demonstrate degraded ecological conditions. This is further supported by earlier studies showing that CPI can be a reliable indicator for prioritizing pollution control in rivers where point and non-point sources co-exist (Sharma and Kansal, 2011). The dominant pollutants contributing to elevated CPI scores such as turbidity, reduced dissolved oxygen, and nutrient enrichment are all indicative of inputs from agricultural runoff, wastewater discharge, and informal waste dumping. These pollutants not only reduce water quality but can also create conditions that enhance the persistence and mobility of microplastics. For instance, elevated organic matter and suspended solids can promote microplastic aggregation, while low oxygen levels may exacerbate stress in aquatic organisms, increasing their susceptibility to ingesting synthetic particles (Wagner *et al.*, 2014).

5.5 Carcinogenic and non-carcinogenic Risk Assessment

Risk assessment of the Ovia River surface water was carried out to evaluate potential human health effects arising from exposure to heavy metals via ingestion and dermal contact. Following standard methodology recommended by the United States Environmental Protection Agency (USEPA, 2004), the chronic daily intake (CDI) values were calculated for each metal. Ingestion emerged as the primary exposure pathway, consistent with findings from other studies on water contamination in tropical river systems (Adekola and Eletta, 2007; Olalekan *et al.*, 2022). Among the metals assessed, essential elements such as iron, zinc, and manganese showed higher levels of intake, likely due to their relative abundance in the aquatic environment. These elevated CDIs may be attributed to both natural geochemical background and contributions from anthropogenic sources such as agriculture, effluent discharge, and land runoff, especially in areas with intense human activities.

Non-carcinogenic risk was assessed through the calculation of hazard quotient (HQ) and hazard index (HI). Although most metals fell within acceptable HQ limits, lead and chromium exhibited higher contributions to the cumulative hazard index, indicating potential health concerns if exposure continues over long periods. More critically, the carcinogenic risk assessment using the incremental lifetime cancer risk (ILCR) model suggested elevated lifetime cancer risks associated with exposure to certain metals, particularly chromium, lead, and nickel. These findings are in line with previous research highlighting the carcinogenic potential of these elements in contaminated surface waters (Ali *et al*., 2019; Rashid *et al*., 2022). ILCR values above established regulatory thresholds point to the possibility of long-term health effects such as organ damage or cancer, especially among vulnerable populations with high water dependence. The elevated risks are particularly concerning in downstream areas where pollutant accumulation is higher due to sedimentation and continuous discharge inputs.

The cumulative ILCR findings emphasize the need for urgent attention to metal contamination in the Ovia River. Although non-carcinogenic risks appear manageable at present, the long-term carcinogenic potential calls for proactive mitigation measures including regular monitoring, pollutant source control, and community awareness. Incorporating microplastic studies alongside heavy metal assessments is also crucial, given the emerging evidence that microplastics can act as vectors for heavy metals, enhancing their bioavailability and toxicity in aquatic environments (Barboza *et al*., 2018). These findings are consistent with previous research. Studies conducted by USEPA (2004) and Ololade *et al*. (2025) in urban water systems have reported similar trends, where ingestion is the primary exposure pathway and elevated ILCR values indicate a heightened carcinogenic risk. The relatively low HI, however, suggests that while the non-carcinogenic risk is manageable, the long-term carcinogenic potential poses a serious concern for public health. Consequently, these results underscore the urgent need for effective water treatment and pollution mitigation strategies to reduce heavy metal contamination in the water supply.

5.6 Contribution to Knowledge

The study has contributed to knowledge in the following ways:

1. The study establishes a comprehensive dataset on microplastic contamination levels in surface water and some fish species from the Ovia River, Benin City, Nigeria.
2. The study revealed significant differences in microplastics ingestion between *Notopterus afer* and *Heterobranchus bidorsalis*, with *H. bidorsalis* exhibiting higher diversity and complexity of microplastics polymers.
3. This study revealed that surface water contained a higher concentration and broader particle size distribution of microplastics compared to the fish species studied

5.7 Conclusion and Recommendation

This study presents the first comprehensive assessment of microplastics contamination in the Ovia River, integrating SEM, FTIR, EDX analyses with physicochemical water quality parameters and health risk assessments. Findings revealed predominantly small, fragmented microplastics in water and fish, reflecting ongoing degradation influenced by environmental factors and local pollution sources. Polymer identification showed water-soluble types, textile fibers, and packaging materials, while elemental analysis detected major elements alongside trace heavy metals such as lead, chromium, and nickel, indicating potential long-term health risks through bioaccumulation. Physicochemical parameters further highlighted moderate pollution levels contributing to microplastic behavior in the river. These results indicate moderate microplastic pollution and associated risks within this tropical freshwater ecosystem, establishing a critical baseline for future research and environmental management. To address these challenges, continuous, multi-seasonal monitoring combining microplastic analysis with physicochemical assessments is essential to track pollutant dynamics. Strengthening waste management including effluent treatment, control of single-use plastics, and public awareness is urgently needed. Health risk models should consider microplastics as vectors for contaminants, and regulatory frameworks must address cumulative pollution effects. Collaboration between researchers, policymakers, and industry will be key to protecting the Ovia River and surrounding communities.

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APPENDICES

Appendix A: Acceptance Criteria for PSD Data for Surface Water

Station	January (Mean > SD?)	March (Mean > SD?)	May (Mean > SD?)
Station 1	✅ (Accepted)	✅ (Accepted)	❌ (Rejected)
Station 2	✅ (Accepted)	❌ (Rejected)	❌ (Rejected)
Station 3	✅ (Accepted)	❌ (Rejected)	❌ (Rejected)
Station 4	✅ (Accepted)	❌ (Rejected)	✅ (Accepted)

Appendix B: Acceptance Criteria for Fish PSD Data

Fish Species	January	March	April
	(Mean > SD?)	(Mean > SD?)	(Mean > SD?)
Notopterus spp	✅ (Accepted)	✅ (Accepted)	❌ (Rejected)
Heterobranchus spp.	✅ (Accepted)	✅ (Accepted)	❌ (Rejected)