

COMPOSITIONAL PROFILES AND SPATIAL DISTRIBUTION OF MICROPLASTICS ACROSS THREE SELECTED RIVERS IN RIVERS STATE, NIGERIA

ABSTRACT

Microplastics (MP) are ubiquitous and persistent contaminants in waterbodies and a pervasive and preventable threat to the health of marine ecosystems. These particles are defined as <5 mm in size and can be introduced into the environment via primary sources such as the use and disposal of microbeads in cosmetic, cleaning products and sandblasting largely contribute, as well as secondary sources which include the fragmentation of litter by mechanical or UV light-induced degradation. This study was conducted to investigate the concentration of microplastic contaminants in Imo River, Ntawogba River and New Calabar River as well as the physicochemical and microbial characteristics of the water bodies. Composite sampling technique was used in this study. The physicochemical analysis of the water samples was carried out both *in-situ* and *ex-situ*. Two techniques (Fourier Transform Infrared Spectroscopy, FTIR and Gas Chromatography-Mass Spectrometry, GC-MS) were used to identify the microplastics and their concentrations. The reference spectra for the FTIR results were in the spectral range stretching from 4000–650 cm⁻¹ while the GC-MS polymer identification library produced results of both qualitative and quantitative assessment of the microplastics. The eight (8) priority MP polymers found in the water samples were polyethylene (PE), polyethylene terephthalate, dioctyl terephthalate, polybrominated diphenyl ether, tetrabromobisphenol A, polypropylene, acrylic fibre, and polystyrene. PE was found to be the most ubiquitous MP (45.74%, 45.84% and 47.76%) across Imo, Ntawogba and New Calabar River, respectively. Results obtained indicate the pervasiveness of MPs in the studied rivers and signify threat to aquatic organisms and man via their biomagnification through the food chain.

Keywords: Microplastics (MP), Fourier Transform Infrared Spectroscopy (FTIR), Gas Chromatography-Mass Spectrometry (GCMS), polyethylene (PE), polyethylene terephthalate, dioctyl terephthalate, polybrominated diphenyl ether, tetrabromobisphenol A, polypropylene, acrylic fibre, and polystyrene.

1. INTRODUCTION

The term "microplastic" (MP) was formally introduced in 2004 by Thompson *et al.* (2004) who alerted to the growing problem of the plastic release to the seas. The production of plastics has increased tremendously in the last 60 years, from approximately 1.7×10^6 tons in the 1950s, to 3.2×10^8 tons in 2015 (Plastics Europe, 2016; Hernandez *et al.*, 2017). This increase was fueled by gradual improvements in plastic manufacturing techniques, which resulted in the production of cost-effective, corrosion-resistant, lightweight, and more durable varieties. Globally, plastics play enormous roles in the delivery of comfort, quality,

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and safety in modern day lifestyles. Consequently, such high plastic production and usage has resulted in increased generation of plastic waste.

Microplastics are plastic debris with its longest diagonal less than or equal to 5mm (Kačicková *et al.*, 2017) and originate from primary and secondary sources. Primary sources fulfill a specific need or function e.g., microbeads used in personal care products, abrasive cleaning particles, pre-production resin pellets, and microfibers used in manufacturing textiles (Peter *et al.*, 2020) whereas, secondary sources result from degradation, wear and tear, or fragmentation of larger debris. Secondary sources include litter fragments (e.g., plastic bags, bottles, wrappers, Styrofoam containers, cigarette filters), synthetic fibers from textiles, road salt, and tire wear particles (GESAMP, 2019; Rødland *et al.*, 2020). In 2015, approximately 6,300 million metric tons of plastic waste was generated, of which only 9% was recycled (Geyer *et al.*, 2017). Plastic debris items, ranging in size from microscopic to macroscopic, have been detected in benthic and pelagic habitats in almost all aquatic ecosystems, including remote locations such as the Arctic and the deep sea (Van Cauwenberghe *et al.*, 2015; Browne *et al.*, 2011). Several human activities potentially introduce microplastic into the aquatic environment, and examples of such activities include the improper disposal of plastics and the intentional use of microscopic plastic particles for personal and industrial uses (e.g., polyester) which are shed during cloth washing (Browne *et al.*, 2007; Cole *et al.*, 2011). Some other sources of microplastics in the aquatic ecosystem include loss of pellets during transportation, wastewater effluent, fishing ropes and gears, cigarette butts, abrasion from sandblasting at shipyards, plastic waste carried by wind or run-off water, and so on (Rochman, 2013).

The impacts posed by microplastic debris depend on the debris size. Large plastic debris such as discarded fishing ropes and nets cause entanglement of invertebrates, birds, and mammals. On the other hand, smaller plastic items, such as bottle caps and plastic pellets, can be ingested, causing obstruction of the gut (Law & Thompson, 2014). The omnipresent nature of microplastics in the environment, poses a serious threat because of its persistence in the environment. They are carriers of some chemicals that are typically found at their highest concentrations in the sea-surface microlayer, which is, of course, the predominant layer of low-density microplastics (Desforages *et al.*, 2014; Foekema *et al.*, 2013). Microplastics have been found to be ingested by some aquatic microorganisms like fishes and other biota and as such, can find its way into the food chain (Schirinziet *et al.*, 2017). The most identified plastic polymers in environmental samples are polyethylene (PE), polypropylene (PP), polystyrene

(PS) and polyvinylchloride (PVC) (GESAMP, 2015) and their preponderance in water bodies in Nigeria, especially in regions with high concentration of industrial activities has not been investigated. Hence, this study was aimed to investigate the concentration of microplastics, and the physicochemical and microbial characteristics of Imo River, Ntawogba River and New Calabar River, in Rivers State, Nigeria.

2. MATERIALS AND METHODS

2.1 Study Area

The study was carried out in Imo River, Ntawogba River and New Calabar River, in Rivers State, Niger-Delta Region of Nigeria. Imo River is in the southern eastern Nigeria and flows 240 kilometres (150 miles) from Onuimo into the Atlantic Ocean. Its estuary is around 40 kilometres (25 miles) wide, (Afigbo- Adiele, 2005) and the river has an annual discharge of 4 cubic kilometers (1.0 cu/ml) (Menally, 1980) with 26,000 hectares of wetland (Afigbo- Adiele, 2005). The source location is Okigwe, Imo state and its coordinates are: $5^{\circ} 50' 56''$ N, $7^{\circ} 14' 20''$ W.

Ntawogba River is a single-channel low gradient freshwater body (Amangbara & Gobo, 2007) which lies on the extreme west of Port Harcourt Metropolis between the approximate longitude $6^{\circ} 58' 00''$ to $7^{\circ} 06' 00''$ E and latitude $4^{\circ} 40' 00''$ to $4^{\circ} 55' 00''$ N (Gobo & Abam, 2006). Its upstream is at Rumueme and Rumuepirikom in Obio-Akpor Local Government Area and flows through Government Reserve Area (GRA) Phase III, a less densely populated area to a more densely populated and high economic activity areas of Diobu Axis of Port Harcourt L.G.A. and eventually empties into Amadi Creek.

New Calabar River and its tributaries are one of the series of low-lying delta river which empty into coastal lagoons and creeks bordering the Atlantic Ocean (Vincent-Akpu & Yanadi, 2014). New Calabar river lies between $4^{\circ} 30' 00''$ and $4^{\circ} 49' 00''$ N and $6^{\circ} 59' 00''$ and $7^{\circ} 00' 00''$ and empties into the Atlantic Ocean (Stanley *et al.*, 2017). The river is freshwater and acidic at the source, but gradually becomes brackish, tidal, and slightly alkaline at the lower zone near its mouth. Figure 1 presents the locations of the studied rivers.

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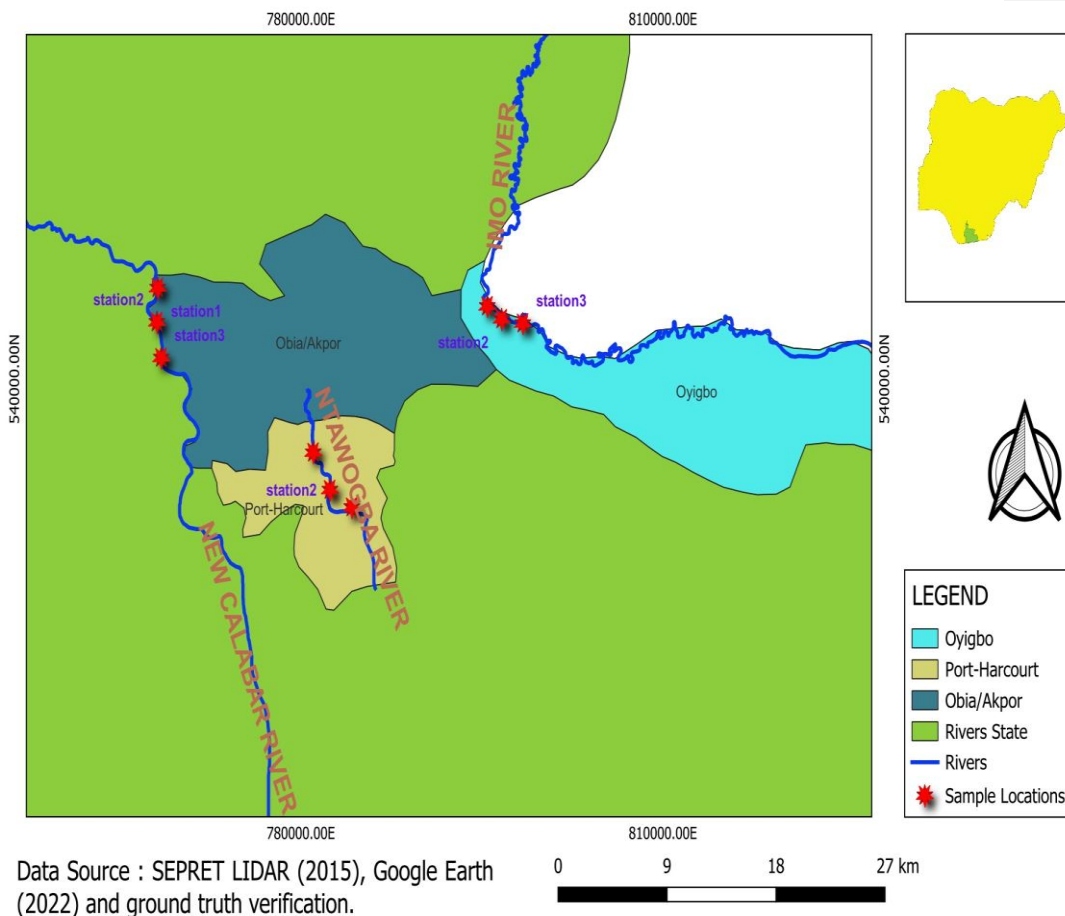


Figure 1. The location of the three rivers studied in Rivers State

2.2 Collection of Water Samples

Using sterile aluminum containers, water samples were collected from three (3) different points in each river. Surface water 1 (SW-1) was collected 100m away from each of the bridges crossing the various rivers: Imo River, Ntawogba River and New Calabar River. Surface water 2 (SW-2) was collected 200m away from SW-1, and surface water 3 (SW-3) was collected 200m away from SW-2 by dipping the sample container 10cm below the water surface against the flow direction. Aluminum sample bottles were conditioned by washing with methanol and allowed to dry under room temperature for 5 days. The pre-cleaned aluminum sampler was immersed 10cm below the water surface and 0.5L of water taken.

Water samples (0.5L) were collected at each sampling location and homogenized to form a composite sample. All sampling points were however geo-referenced, and the coordinates are as presented in Table 1. At each sampling site, the aluminum sampler was rinsed three times before sample collection. Samples for different parameters were taken and preserved according to the method of APHA (2005) and Aiyesanmi (2006) to ensure accuracy and reliability of results and implementing quality control measures to minimize contamination during water sample analysis at the laboratory.

Table 1: GPS coordinates of the different sampling stations

	Imo River	Ntawogba River	New Calabar River
SW-1	7° 8' 44" N 4° 53' 19" E	4° 48' 33" N 7° 00' 56" E	4° 53' 19" N 6° 53' 53" E
SW-2	4° 53' 17" N 7° 8' 45" E	4° 48' 32" N 7° 00' 56" E	4° 53' 20" N 6° 53' 54" E
SW-3	4° 53' 15" N 7° 8' 46" E	4° 48' 31" N 7° 00' 56" E	4° 53' 21" N 6° 53' 55" E

2.3 Analysis of Samples

2.3.1 Physicochemical Analysis

The Hanna Multi-parameter water checker was used to determine pH, temperature, electrical conductivity, turbidity, dissolved oxygen, salinity, and dissolved solutes in the water samples. The probe was first calibrated, then dipped into each sample and the displayed reading was allowed to stabilize before recording. The temperature of the sample was measured in degree Celsius while the electrical conductivity of samples was measured in milliSiemens per meter (mS/m). The turbidity of the samples was measured and reported in NTU (Nephelometric Turbidity Units). Dissolved oxygen, salinity and dissolved solids of the water samples were measured in milligrams per litre (mg/L).

The APHA 2540D test method was used for the determination of total suspended solids (TSS). Deviation from the method was that Whatman filter paper (0.45 µm) was used instead of glass fibre filter and the TSS content was calculated using Equation (1):

$$TSS \left(\frac{mg}{L} \right) = \frac{(A - B)1000}{Sample Volume (mL)} \quad (1)$$

Where A is the weight of filter paper + residue (mg) and B is the weight of filter paper (mg).

APHA 5220 D/ HACH 8000 method was employed for the determination of chemical oxygen demand (COD) and the COD results were defined as the mg of O₂ consumed per litre of sample under conditions of this procedure. The 5-day BOD test method (APHA 5210B) was used for BOD₅ determination. Samples for the BOD₅ test were incubated in the dark for 5 days at 20°C. The residual dissolved oxygen was determined electrometrically after the incubation period and the BOD₅ calculated afterwards. APHA 4500-Cl⁻ B test method was used in the titrimetric determination of the concentration of chloride. In a neutral or slightly alkaline solution, with potassium chromate as indicator for the end point of silver nitrate titration of chloride, silver chloride is precipitated quantitatively before red silver chromate is formed.

2.3.2 Microbiological Analysis

Counts of total heterotrophic bacteria and fungi in the water samples were determined using APHA 9215B/9610B and ASTM D 5465-93 (pour-plate) test methods. Serial dilution of the water samples was carried out using sterile normal saline. Aliquots (1mL) of the 10-fold dilutions were plated on nutrient agar and Sabouraud dextrose agar for the enumeration of heterotrophic bacteria and fungi, respectively. Bacterial plates were incubated at 35±2°C for 24-48 hours whereas fungal plates were incubated at 25±2°C for 3-5 days. Counts of microorganisms in samples were presented as colony-forming units per millilitre (cfu/mL) of water sample.

Counts of hydrocarbon utilizing bacteria were determined using APHA 9215C/ASTM 5465-93 (spread plate) test method. Serial dilution of the water samples was carried out using sterile normal saline. Aliquots (0.1mL) of the 10-fold dilutions were spread on minimal medium containing the appropriate mineral salts for bacterial growth. Crude oil-moistened filter papers placed on the lid of the inverted plates provided the carbon source for growth. The plates were incubated at 25±2°C (for bacteria & fungi) for 7-10 days. After incubation, colonies that developed were counted and presented as cfu/mL of water samples.

2.3.3 Concentration of Microplastics in Water Samples

A Fourier transform infrared (FT-IR) spectroscopy was used for identification of the polar functional groups of the microplastics polymer. The samples were subjected to contact with

infrared (IR) radiation. The IR radiations then had impacts on the atomic vibrations of molecules in the sample. The wavelength of incident laser was set to 532nm, and the FT-IR spectra were from 650 to 4000cm⁻¹. The FT-IR images of the samples were analyzed to identify and quantify the functional groups of the microplastics present.

Gas Chromatography-Mass Spectrometry (GC-MS) analysis was carried out using an Agilent 6890 gas chromatograph with a 5973 MS detector equipped with 30-m x 0.25-mm and 0.32-mm ID fused-silica capillary column chemically bonded with SE-54 (DB-5 or equivalent), and 1- μ m film thickness. (Agilent). The following temperature ramp was used: injector at 250°C, oven initially at 200°C, held for 1 min and heated to 230°C (1.5°C min⁻¹, then held for 10min). Helium was used as the carrier gas at a flow rate of 1 mL min⁻¹. The split ratio was 50:1, and the sample size was 2 μ L. The characterization and identification of the microplastics, from the sample was completed in the thermo Excalibur 2.1 0.114 acquisition software.

In Scanning Electron Microscopy (SEM) imaging, microplastics (MP) were isolated from the water samples by filtration through a fine mesh filter of 0.45 μ m pore size. These MPs were cleaned to remove adhering debris that could interfere with imaging, prepared, and coated for imaging and then transferred onto a suitable sample holder. It was ensured that the MPs were completely dry to prevent water vapour from affecting the SEM imaging process. The prepared sample stub was loaded into the SEM chamber, ensuring it was properly positioned to prevent electron scattering. The appropriate electron beam was set in accelerating voltage and current based on the nature of MPs and the working distance was adjusted to optimize the focus of the electron beam on the sample surface. Afterwards, SEM images of the MPs were acquired at various magnifications to capture their surface features, shapes, and sizes. Different imaging parameters were experimented to achieve the best contrast and resolution.

2.4 Statistical Analysis

The results were subjected to descriptive analysis and Pearson's correlation. Pearson's correlation was used to assess the relationship between the microplastics, and some physicochemical properties of the water samples collected.

3. RESULTS AND DISCUSSION

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Water has a neutral pH of 7 which indicates that it is neither acidic nor basic where scale ranges from 0 (very acidic) to 14 (very alkaline). The results in Table 2 showed that New Calabar River with a mean pH of 5.66 was slightly acidic compared to Imo River and Ntawogba River with mean pH values of 6.57 and 6.44 respectively which are closer to the pH neutral value of 7 for water. The U.S. Environmental Protection Agency (EPA) recommends that the pH of water sources should be between 6.5 to 8.5. Going with EPA standard, water source from New Calabar River is unfit for domestic use unless it undergoes further treatment to take care of the acidity. The results in this study agreed with the report of Nzeako *et al.* (2014) in their assessment of freshwater body in Niger-Delta, Nigeria with pH of 5.15 to 5.82. This acidic nature of New Calabar River is probably due to industrial discharges, illegal sand mining, burning of tyres to roast cow-skin, and direct refuse disposal into the water body. Water with pH outside the normal range (6.5 to 8.5) can adversely affect the growth and development of aquatic life (Kalagboret *et al.*, 2020).

Salinity of surface water is relatively uniform as it is generally well mixed by waves, wind, and tides (Chikwe, 2020). It is an important factor in understanding the properties and characteristics of waterbodies. For this study, New Calabar River had a mean salinity of 0.44mg/L (Table 2) which agreed with the report of Chikwe (2020) in her assessment of pollution status of Imo River, Southeastern Nigeria with mean salinity of 0.48mg/L in Asa sampling station, during dry season. This could be attributed to the human activities there such as industrial discharge, and illegal sand mining.

The mean conductivity value of New Calabar River is 905.33 μ S/cm (Table 2) and slightly exceeded the WHO recommended standard. WHO requirement for aquatic life is 900 μ S/cm. Since conductivity is a useful indicator of mineralization and salinity in water, this could be a reason why salinity value (Table 2) is also highest in New Calabar River compared to the other rivers studied. Conductivity qualitatively reflects the status of inorganic pollution and is a measure of total dissolved solid and ionized species in the water (Chikwe, 2020).

Ntawogba River with a COD mean value of 75.67mg/L (Table 2) was highest amongst the studied rivers. This could be attributed to its lentic nature, indiscriminate refuse dumping of

wastes, open defecation, and construction activities happening around this waterbody. This trend was also observed by Ogunfowakan *et al.* (2005) in their study. The values obtained in this study are also similar with those reported by Amadi (2010), in his study of water quality indices of Otamiri and Oramiriukwa Rivers. High COD values suggest the presence of a significant amount of organic matter in a waterbody, which can include fecal matter, sewage, industrial effluents, and agricultural runoff.

From Table 2, chloride concentrations for the rivers studied met the WHO recommended limit of 250mg/L for aquatic life. New Calabar River with a mean chloride value of 230.33mg/L had the highest chloride concentration which gave credence to the benign salinity of the water samples.

The mean BOD value as stated in Table 2 for Imo River was 8.25mg/L and 33.50mg/L for Ntawogba River while New Calabar River was 15.67mg/L. Although these values are in consonance with the report of Chindah *et al.* (2011), they are higher than WHO recommended standard of 5.0mg/L. Ntawogba River showed the highest mean BOD value (33.5mg/L), which could be likely attributed to the presence of a significant amount of readily biodegradable organic matter in the river which indicated pollution from sources like untreated sewage, industrial effluents, and agricultural runoff. Oxygen is required for respiration by microorganisms involved in the decomposition of these organic materials (Nartey *et al.*, 2012).

THB were found in high numbers in all the water samples analyzed (Table 2) and was expressed in colony-forming units per millilitre (CFU/mL) of water sample. Mean THB count was highest in Ntawogba River (1.29×10^5 CFU/mL) when compared to the other rivers studied. Likewise, mean HUB count was highest (1.10×10^3 CFU/mL) in Ntawogba River (Table 2) indicating the probable contamination of the water by petroleum hydrocarbons. HUBs proliferate in environments contaminated with petroleum hydrocarbons since they possess the metabolic capability for assimilation and utilization of these compounds as sources of carbon and energy. Hence, they are important players in the natural breakdown of hydrocarbon pollutants and restoration of polluted environments. Several anthropogenic activities occurring around the Ntawogba river could have predisposed the river to hydrocarbon pollution.

Figs. 2 – 4 indicate the concentrations of MPs in the three rivers in Port Harcourt studied. MPs were detected in all water samples from the rivers studied with polyethylene (PE) being the predominant microplastic type in the surface water bodies with average abundance of 45.91%, 40.18%, 44.40% across the Imo River, Ntawogba River and New Calabar River

respectively. Polyethylene terephthalate (PET) was next to PE in predominance as indicated on the GC-MS chromatograms (Figs. 5–7). This phenomenon can be linked to the vast consumption of plastic products (e.g., single-use plastics), littering and poor waste management culture which result in microplastics ending up in the waterbodies via storm water runoff or wind. Among the microplastics detected in the rivers, tetrabromobisphenol A (TBA) and polybrominated diphenyl ether (PBDE) were the least in magnitude obtained. Both forms of plastic are used as flame retardant coatings of electronic devices such as televisions, mobile phones, and computers and become e-waste at the end of their life cycle. Most of these devices end up in landfills which could be the reason why their concentrations were low in surface water of the three rivers.

Figs. 8–10 are graphical representation of the FTIR spectrum, highlighting the absorption peaks of interest. Here, the prominent absorption peak at 2920cm^{-1} corresponds to aliphatic C-H (Hydrocarbon) stretching, indicating the presence of CH_2 (Alkene) and CH_3 (Alkane) groups in the microplastic samples. The peak at 1725cm^{-1} is associated with carbonyl (C=O) stretching, suggesting the presence of polymers with carbonyl functional groups. The band at 1600cm^{-1} is a characteristic of aromatic C=C stretching, indicating the presence of aromatic polymers. The absorption in the $1200\text{--}1000\text{cm}^{-1}$ range suggests C-O stretching vibrations which can be indicative of polyester and polyethylene plastics. The region between 800 and 700cm^{-1} corresponds to out-of-plane bending vibrations commonly seen in aromatic groups.

The SEM images presented here (Figs. 11–13) provide a close-up view of the microstructures and offer a glimpse into the microscopic world of the identified MPs, showcasing their unique structures and surface properties.

Physicochemical properties of the water samples may provide insights into the conditions that influence the presence and transport of MPs. The increase in salinity may enhance the MP adsorption capacity for organic pollutants by causing a change on the strength of electrostatic interaction and the degree of ion exchange in the adsorption process (Gao *et al.*, 2023). As shown in Table 2, salinity level was highest in New Calabar River (NCR) with a mean value of 0.44mg/L ; the probable reason why polyethylene obtained from New Calabar River was also highest at 44.40%. The aggregation behaviours of MPs regulate their fate and ecological risks in aquatic environments, and it is described that attachment efficiency of heteroaggregation depends on surface interaction of aggregating particles, characteristics of plastic particles, and features of surrounding medium such as pH, natural organic matter, and ionic strength (Atugoga, *et al.*, 2021). However, these phenomena were not investigated in this study.

MPs have not only caused physical damage to marine organisms, but they also introduce potential hazards that have had a negative impact on marine ecosystems (Gao *et al.*, 2023). Fishes mostly (80%) ingest blue PE fragments because the colour and size are more like copepods that they usually consume which causes reduced body weight, growth inhibition, impairment of the reproductive systems, reduced mobility, and finally mortality (Atugoga, *etal.*, 2021).

However, mitigation of MPs in water bodies is a complex challenge that requires painstaking strategies at various levels, from individual actions to policy changes. It is a collective effort involving individuals, communities, industries, government, and the scientific community. By taking proactive steps to reduce plastic use, improve waste management, and advocate for policy changes, it is possible to work towards healthier and more sustainable aquatic ecosystems.

Table 2: Physicochemical characteristics of surface water samples obtained from Imo River (IM-RIV), Ntawogba River (NTW) and New Calabar River (NCR)

Parameter	Imo River	NtawogbaRiver	New Calabar River	NESREA Limits	USEPA Limits
pH	6.57±0.17	6.44±0.05	5.66±0.04	6.5–8.5	6.5–9
Conductivity(µS/cm)	12.00±5.29	397.33±72.47	905.33±174.00	–	1000
Total Dissolved Solids (mg/L)	6.00±2.65	197.33±35.64	453.00±87.00	–	500
Turbidity(NTU)	41.70±2.55	15.97±1.18	19.17±8.84	–	1
Dissolved Oxygen (mg/L)	7.22±0.46	4.07±0.38	5.48±0.14	6.0	0.05
Salinity(mg/L)	0.01±0.00	0.19±0.04	0.44±0.09	–	–
Temperature(°C)	27.93±0.25	29.97±0.72	29.60±0.17	–	–
Redox Potential(mv)	149.67±26.27	75.73±3.50	143.67±1.53	–	–
Chloride (mg/L)	1.26±0.22	103.33±19.55	230.33±70.85	300	250
BiochemicalOxygen Demand (BOD) (mg/L)	8.25±0.12	33.50±1.78	15.67±0.60	3.0	–
Chemical Oxygen Demand	23.67±1.53	75.67±3.06	33.67±1.53	30	250

(mg/L)						
Total Solids	Suspended	21.33±1.53	31.33±2.52	26.33±1.53	0.25	—
Grease(mg/L)						
Total Oil and Grease		0.64±0.11	1.24±0.26	0.47±0.04	0.01	—
THB (CFU/mL)						
		15900.00±14381.58	129333.33±21007.94	16666.67±5783.02	—	—
HUB (CFU/mL)						
		170.00±206.64	1103.33±105.04	370.33±203.81	—	—
THF (CFU/mL)						
		680.00±589.24	10366.67±635.09	1.00±0.00	—	—
HUF (CFU/mL)						
		133.33±57.74	1.00±0.00	1.00±0.00	—	—

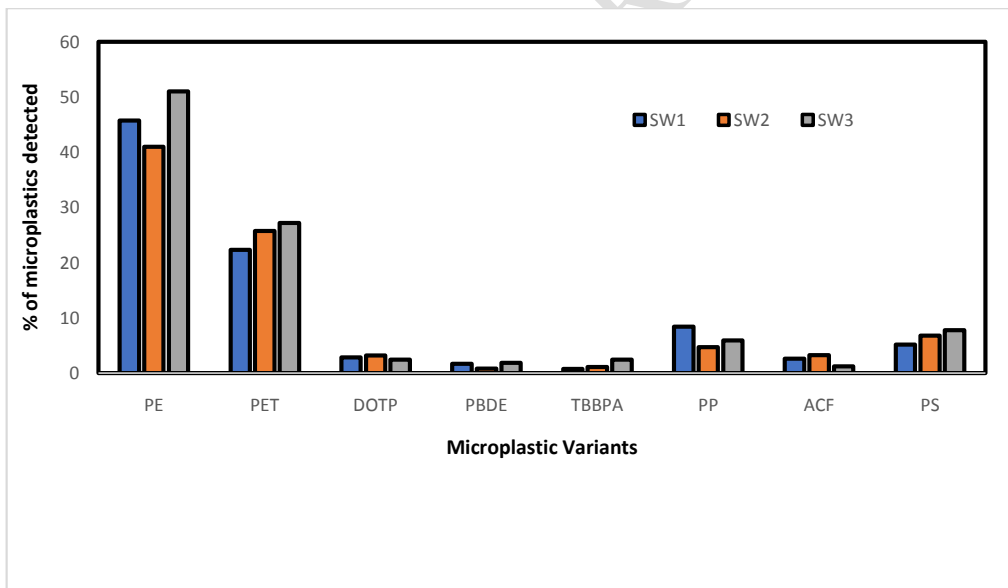


Figure 2: Microplastic concentration (in percentage %) of water samples collected from Imo River

LEGEND:

PE – Polyethylene

PET – Polyethylene Terephthalate

DOTP – Dioctyl Terephthalate

PBDE – Polybrominated Diphenyl Ether

TBBPA – Tetrabromobisphenol A

PP – Polypropylene

ACF – Acrylic Fiber

PS – Polystyrene

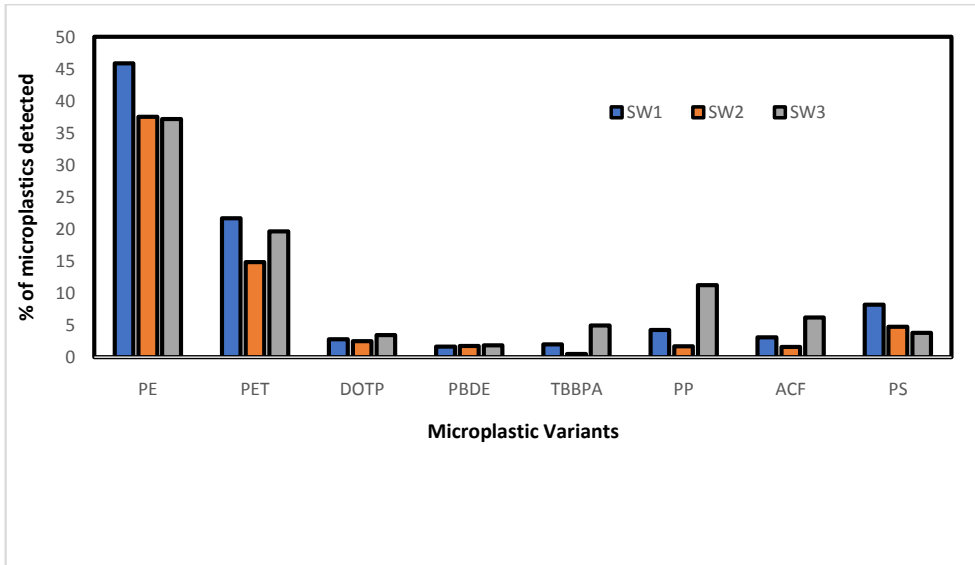


Figure 3: Microplastic concentration (in percentage %) of water samples collected from NtawogbaRiver

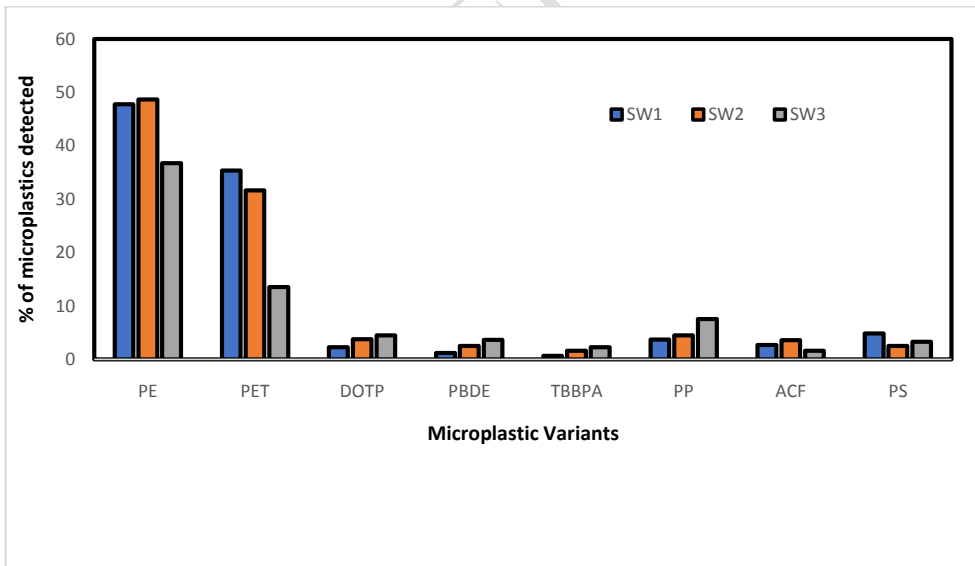


Figure 4: Microplastic concentration (in percentage %) of water samples collected from New Calabar River

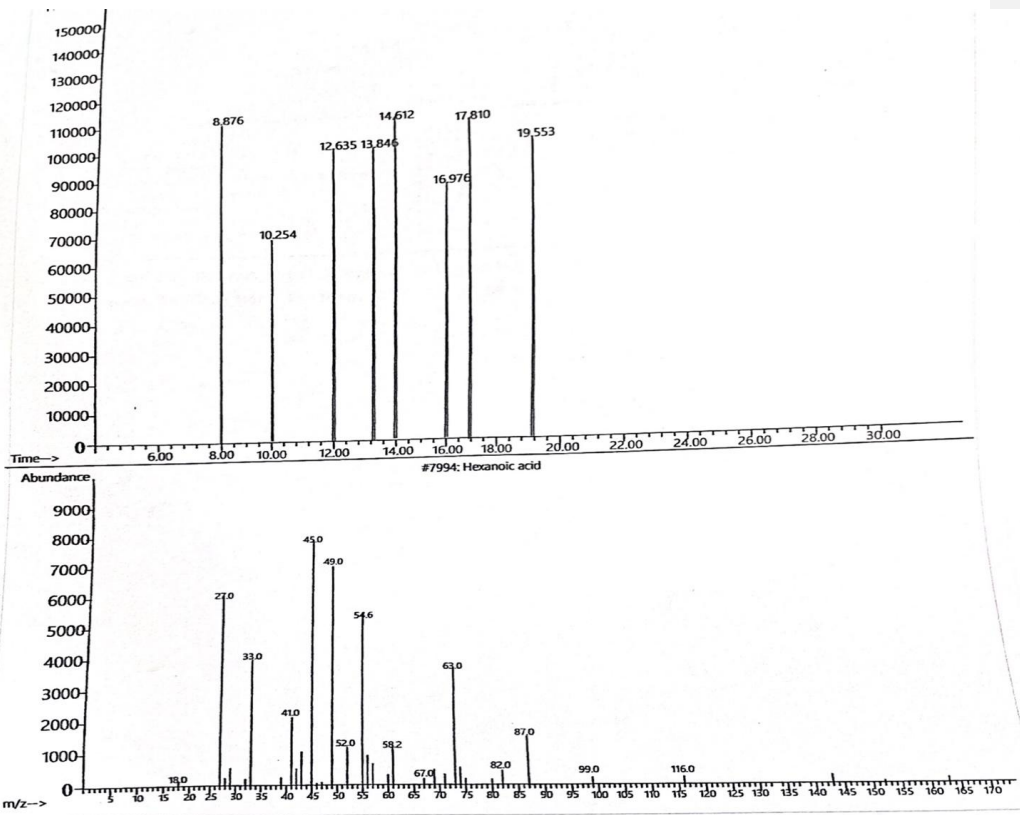


Figure5: GC-MS chromatogram of water samples obtained from New Calabar River (SW1)

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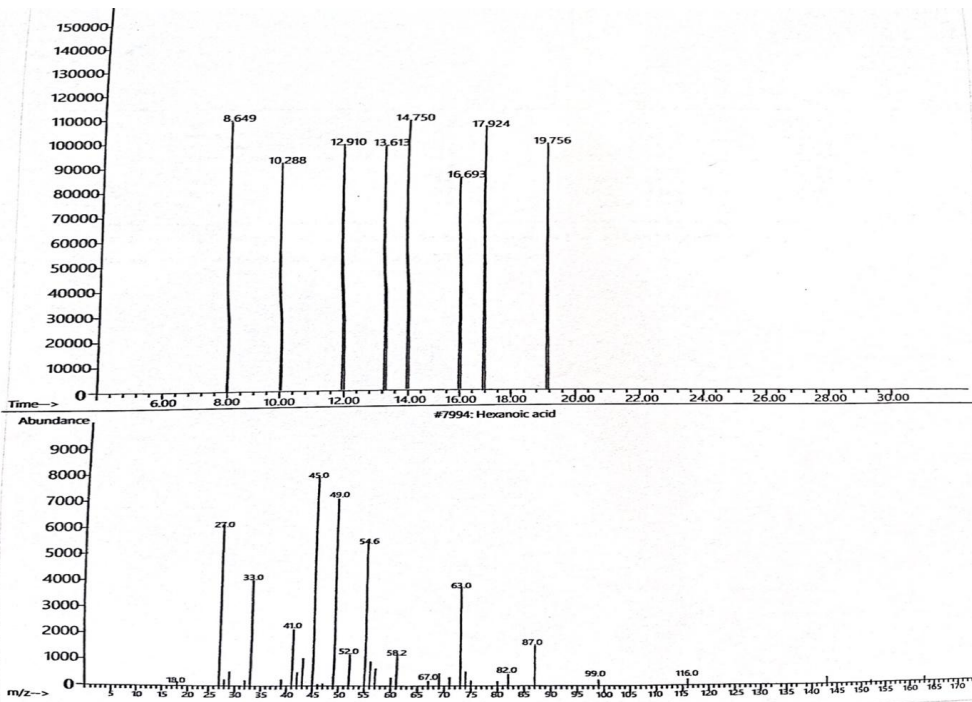


Figure6: GC-MS chromatogram of water samples obtained from Imo River (SW1)

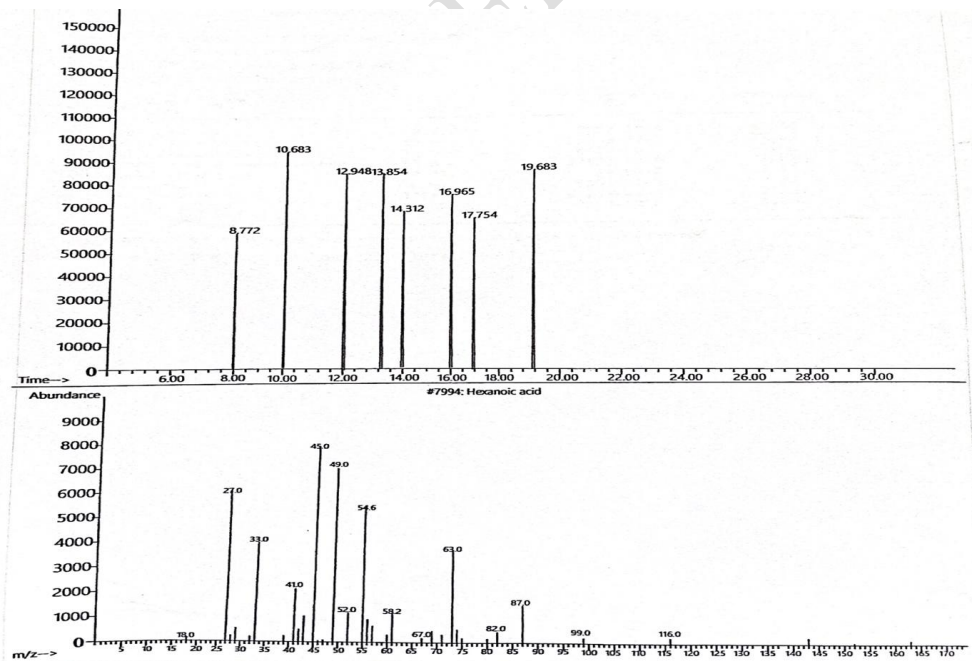


Figure7: GC-MS chromatogram of water samples obtained from Ntawogba River (SW1)

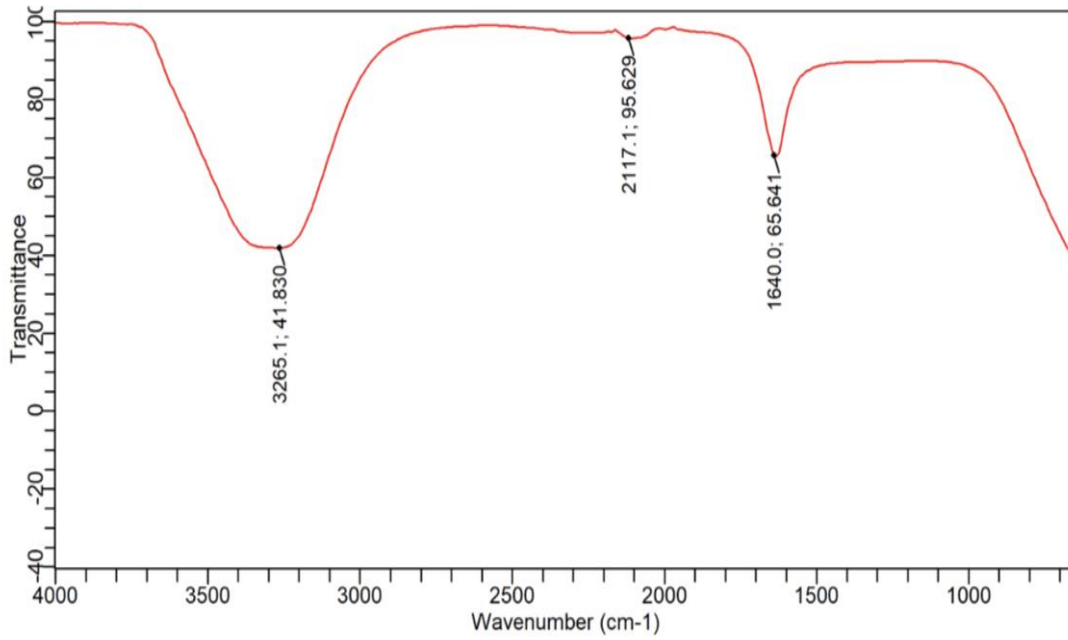


Figure8: FT-IR spectra of microplastics obtained from New Calabar River (SW1)

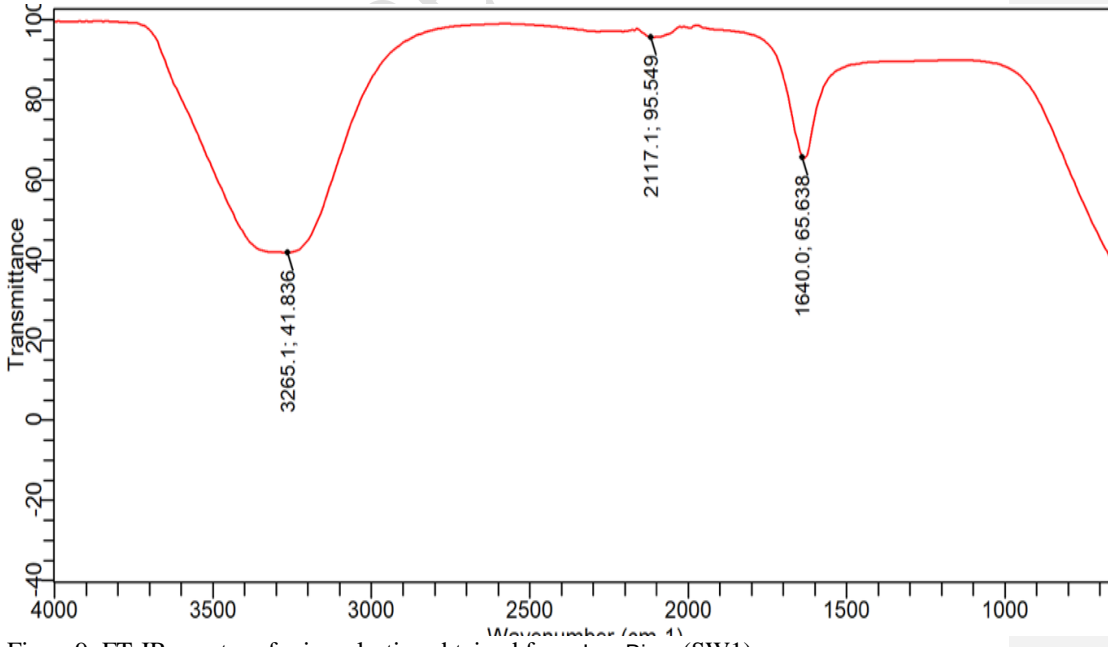


Figure9: FT-IR spectra of microplastics obtained from Imo River (SW1)

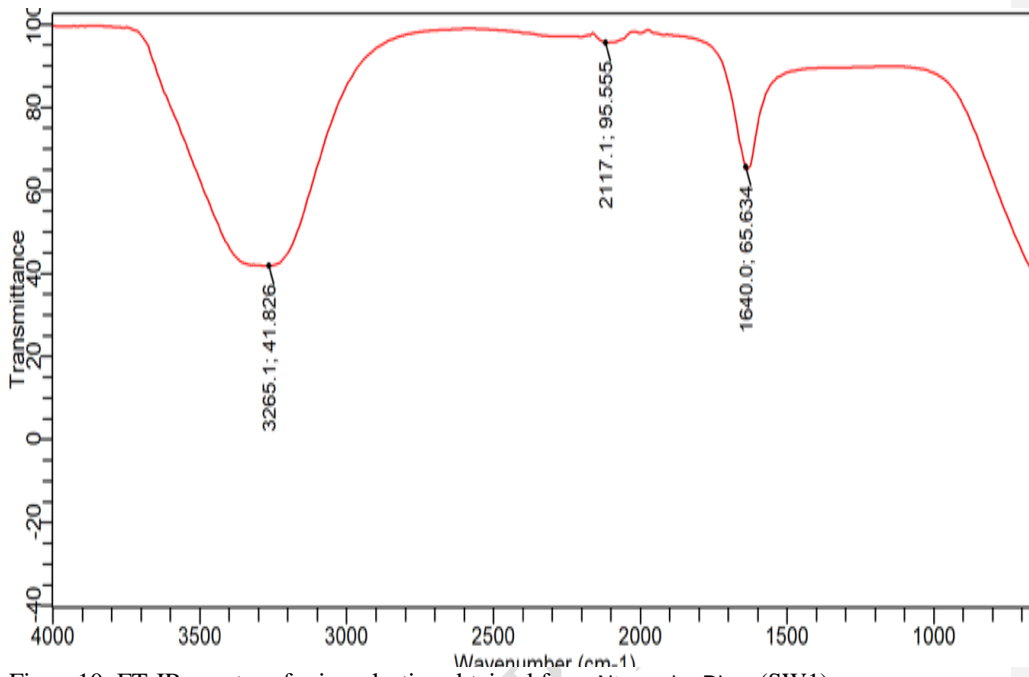
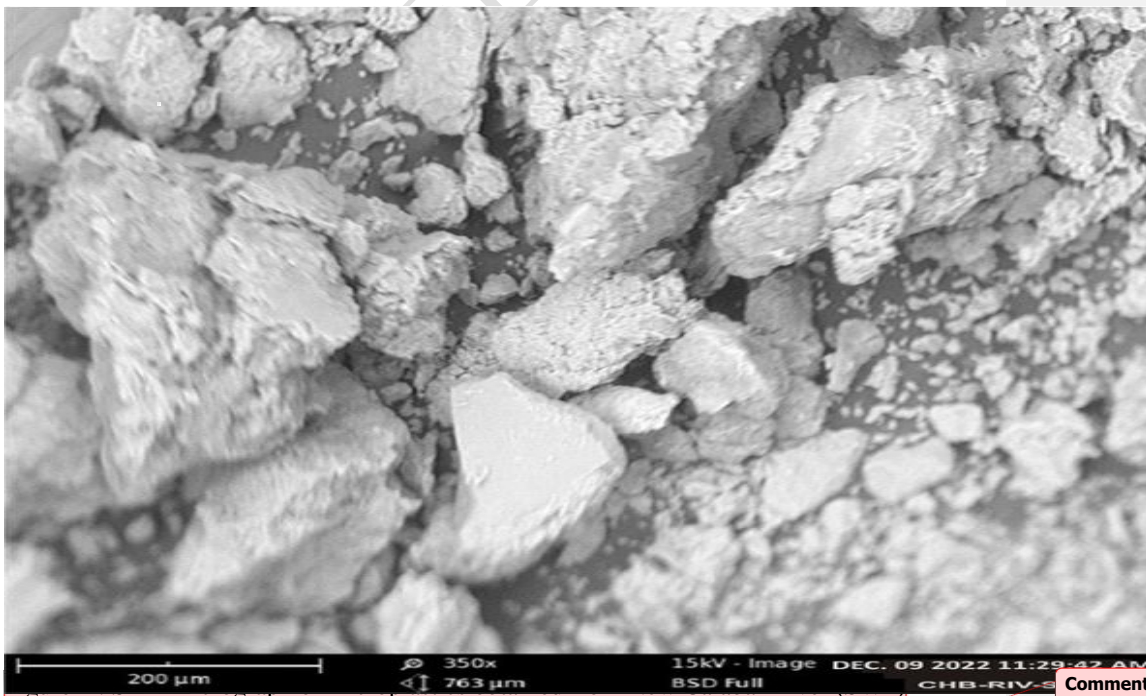


Figure10: FT-IR spectra of microplastics obtained from Ntawogba River (SW1)



Comment [U7]: The SEM test is not explained in the method, if it is carried out the results must be described and explained in detail and discussed

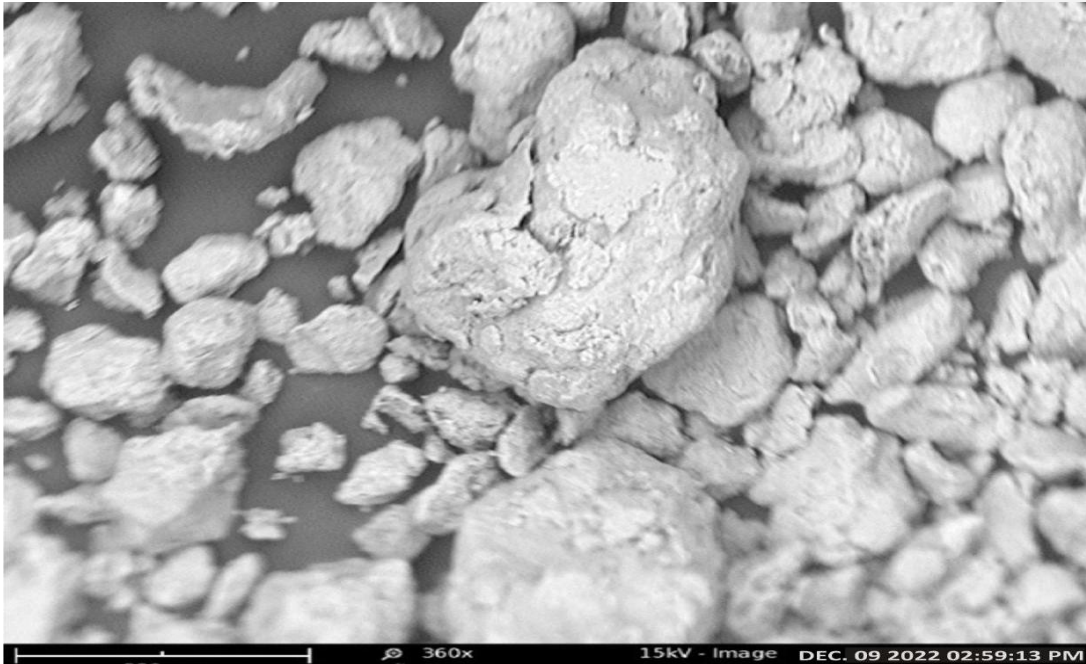


Figure12: SEM micrograph of microplastics obtained from Imo River (SW1)

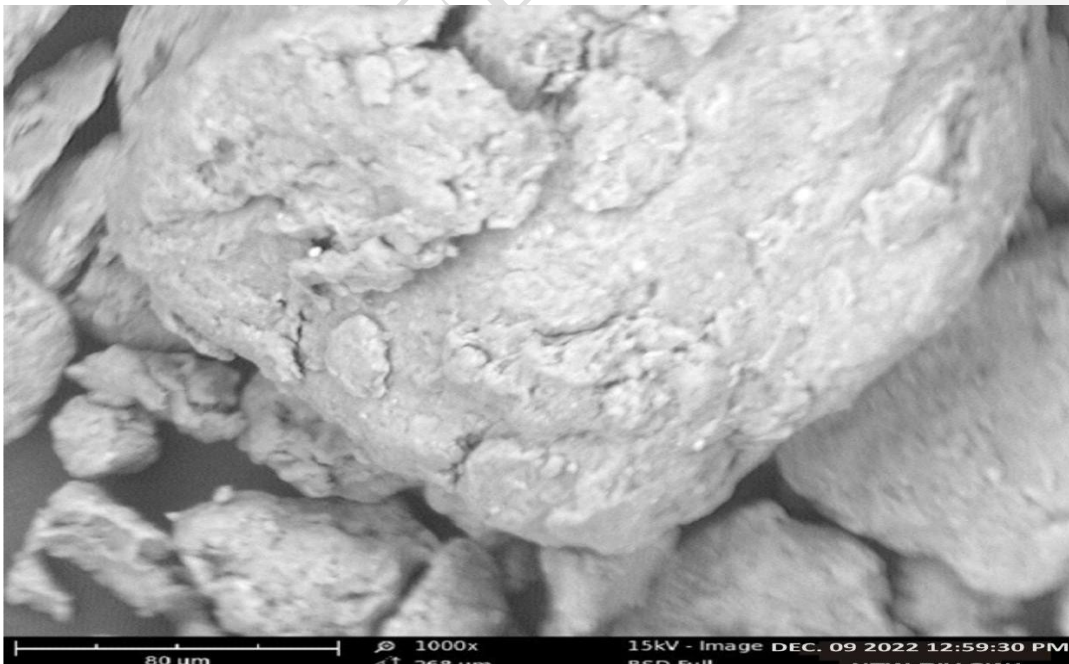


Figure13: SEM micrograph of microplastics obtained from Ntawogba River (SW1)

4. **Conclusion**

Comment [U8]: too long, it's best to summarize i

This study focused on investigating the compositional profiles and spatial distribution of microplastics across three selected rivers viz: Imo, Ntawogba, and New Calabar River in Rivers State, Nigeria utilizing Gas Chromatography-Mass Spectrometry (GC-MS), Fourier Transform Infrared Spectroscopy (FTIR), and Scanning Electron Microscope (SEM) imaging alongside assessing some of the physicochemical and microbiological parameters of the selected rivers. MPs were detected in all water samples from the rivers studied with polyethylene (PE) being the predominant microplastic type in the surface water bodies with average abundance of 45.91%, 40.18%, 44.40% across the Imo River, Ntawogba River and New Calabar River respectively. Polyethylene terephthalate was next to PE in predominance. Among the microplastics detected in the rivers, tetrabromobisphenol A (TBA) and polybrominated diphenyl ether (PBDE) were the least in magnitude obtained. The FTIR spectra corresponded to aliphatic C-H stretching, indicating the presence of CH₂ and CH₃ groups in the microplastic samples, carbonyl (C=O) stretching, suggesting the presence of polymers with carbonyl functional groups, aromatic C=C stretching, indicating the presence of aromatic polymers, and C-O stretching vibrations which indicated polyester and polyethylene plastics presence. The SEM images provided a close-up view of the microstructures and surface characteristics of each microplastic.

This interdisciplinary study holds substantial significance for understanding the overall health of freshwater ecosystems, the load of microplastics, and possible sources of contamination. Therefore, the results in this study may guide environmental conservation efforts, policy formulation, and freshwater resource management in Rivers State, Nigeria.

8. **Life Science Reporting**

No life science threat was practiced in this research.

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Comment [U9]: Reference writing must be consistent, follow established guidelines

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