CSIR COLLEGE OF SCIENCE AND TECHNOLOGY

OCCURRENCE OF MICROPLASTICS IN WATER, SEDIMENT, CULTURED AND WILD FISH IN THE LOWER VOLTA BASIN OF GHANA

YAA ASABEA AGADZI

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BY

YAA ASABEA AGADZI

Thesis submitted to the Department of Fisheries Science and Aquaculture of CSIR College of Science and Technology, in partial fulfilment of the requirements for the award of Master of Philosophy degree in Aquaculture

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DECLARATION

Candidate's Declaration

I hereby declare that this thesis is the result of my own original research and that no part of it has been presented for another degree in this College or elsewhere.

Candidate's Signature:………………………..........Date…………….............

Name: Yaa Asabea Agadzi

Supervisors' Declaration

We hereby declare that the preparation and presentation of the thesis were supervised in accordance with the guidelines on supervision of thesis laid down by the CSIR College of Science and Technology.

Principal Supervisor's Signature ……………………......Date:…………….......

Name: Dr. Ruby Asmah

Co-supervisor's Signature:……………………………Date……………..........

Name: Dr. Pennante Naa Ayikailey Bruce-Vanderpuije

ABSTRACT

Despite the recent surge in microplastic research in Ghana, there is limited information on MPs in the Lower Volta Basin- an important freshwater resource that provides fish for human consumption. This study aims to fill in knowledge gaps by providing baseline information on MPs sampled from cultured and wild fish species, river water and sediment from Asikuma Labolabo, Kpong, and Sogakope Agodormi along the Lower Volta Basin, during the wet and dry seasons. The fish species analysed included *Oreochromis niloticus* from aquaculture farms and 12 local species- *Chrysichthys nigrodigitatus*, *Chrysicthys auratus*, *Tilapia guineensis*, *Tilapia zilli*, *Mormyrus* spp, *Synodontis vellifer*, *Synodontis schall*, *Parachanna obscura*, *Auchenoglanis occidentalis*, *Brycinus brevis* and *Malapterurus electricus* sampled from the wild. The overall percentage abundance of MPs in both cultured and wild sites, for both seasons, was highest in Asikuma Labolabo (47 %), followed by Kpong (33 %), and Sogakope Agodormi (20 %); there were significant differences in MP abundance obtained from the three sampling sites/locations ($p < 0.05$). The total microplastic concentration obtained in fish [wet =1586 MP items/fish, dry = 1114 MP items/fish] in the wet season was higher than for the dry season. Mean microplastic concentrations was higher in sediment (97.50 ± 78.78 MP items/g sediment) in comparison to water (11.10 ± 10.61 MP items/L water). All three different brands of fish feed used at various aquaculture farms, for fish production, recorded mean levels of MPs (A_{feed} = 25.00 + 13.57 items/g, B_{feed} = 15.67 ± 6.15 items/g, and C_{feed} = 6.67 ± 3.85 items/g). Within the matrices of the Lower Volta Basin, fibres were the dominant MP type identified.

Results also indicated that Ghanaian adults who consume freshwater fish from the lower Volta basin are exposed to MP concentrations between 287.10 and 2595.46 MP gram person^{-1} year^{-1}. This study recommends the use of biodegradable alternatives such as bamboo and rafia in place of plastics as cage materials in aquaculture.

KEY WORDS

Cultured and wild fish

Human exposure

Lower Volta Basin

Microplastics

Sediment

Water

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DEDICATION

To Dr. Seth K. Agyakwah, Mr. Bacon Atsu Amengor and my family

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LIST OF ACRONYMS

ALDFG	–	Abandoned, Lost or otherwise Discarded Fishing Gears
ANOVA	–	Analysis of Variance
FAO	–	Food and Agricultural Organization
FM	–	Fish Meal
GDP	–	Gross Domestic Product
MPs	–	Microplastics
PA	–	Polyamide
PBDEs	–	Polybrominated diphenyl ethers
PBTs	–	Persistent Bioaccumulative Toxic Substances
PCA	–	Principal Component Analysis
PE	–	Polyethylene
PET	–	Polyethylene terephalate
POPs	–	Persistent organic pollutants
PP	–	Polypropylene
PS	–	Polystyrene
PVC	–	Polyvinyl chloride
SME	–	Small and Medium-sized Enterprise
WWTP	–	Wastewater Treatment Plants

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CHAPTER ONE

INTRODUCTION

The aim of the introductory chapter is to generate a general context for the study. The chapter states the background to the study, the problem statement research questions and significance of the study.

Background to the Study

Microplastics (MPs) are defined as small-sized plastic fragments and particles that are less than 5 mm in their longest dimension ((Lusher, Hollman, & Mendoza, 2017). MPs can be categorized as either primary or secondary based on their source (Parker, Andreou, Green, & Britton, 2021)

Primary MPs are deliberately manufactured within this general size range for use in industry or various cosmetic products. Primary MPs include pre-production resin pellets, industrial scrubbers for abrasive blast cleaning, and capsules and microbeads in personal care products (Eerkes-Medrano &Thompson, 2018). On the other hand, secondary MPs are formed from the breakdown of larger plastics through physical, chemical and biological degradation. Examples of secondary microplastics include fragments arising from plastic bottles, plastic bags, and other packaging materials (Eerkes-Medrano & Thompson, Parker *et al*;(2012).

Microplastics are heterogeneous mixtures of differently shaped materials referred to as fragments, fibres/filaments, beads/spheres, films/sheets, pellets, amongst others (Lusher *et al.*, 2017). Owing to their varied types and wide range of sources, MPs consist of a wide spectrum of polymers which include: polyethylene (PE), polyethylene terephthalate (PET), polystyrene (PS), polypropylene (PP), high-density polyethylene (HDPE), polyvinyl chloride (PVC), and polyurethane (PU) (Mendoza & Balcer, 2019). Microplastics have high mobility (float, sink) and long periods of dwell time (settle), and can enter into the environment through discharge of wastewater plants for treatment, weathering and degradation of plastic waste in waterbodies and soil erosion or runoff from terrestrial inputs (Acquah, Liu, Hao, Ling, & Ji, 2021). In the aquatic environment, MPs can end up on beaches, sediments, surface waters and within the water column (Shruti, Jonathan, Rodriguez-espinosa, & Rodríguez-gonzález, 2019).

Environmental scientists started investigating the issue of microplastics since the early 2000s (Wagner *et al.*, 2014); it has become an issue of increased concern because of its widespread presence in the aquatic environment and presents potential physical and toxicological risks to organisms (Thompson, 2019). In the aquatic environment, studies have shown that the Arctic Ocean surface waters hold the most plastics of any ocean basin. The number of particles measured in some parts of the bottom of the Arctic Ocean are the highest in the world (Katzc, 2019). Reviews show that, globally, most of the studies on MPs in freshwater have been conducted by Asian countries, especially China (Sarijan, Azman, Ismid, Said, & Jamal, 2020). Thus, Chinese rivers account for about two-thirds of the plastics in the ocean. The Taihu Lake located in the developed areas of China, and closely related to human activities, is reported to have its water and sediment heavily polluted.

Microplastics contain a mixture of chemicals added during manufacturing; the so-called additives efficiently sorb (adsorb or absorb) persistent, bioaccumulative and toxic substances (PBTs) from the environment (Wagner & Lambert, 2018). The small size of microplastics results in their uptake by a wide range of aquatic species; this disturbs their physiological functions, which then go through the food web creating adverse health issues in human (Issac & Kandasubramanian, 2021). In aquatic ecosystems, primary productivity can be affected when MPs adhere to the surface of algae and other vascular plants and cause reduction in their photosynthetic efficiency (Ge *et al*., 2021). The ingestion of microplastics by aquatic organisms including species of commercial importance for fisheries and aquaculture, and the accumulation of PBTs have been central to the perceived hazard and risk of microplastics in the marine environment (Wagner & Lambert, 2018). This negatively affects populations of wild and farmed aquatic organisms, with an adverse effect on food security. Ingestion of contaminated fish, consumption of microplastic-contaminated tap water, sea salt and bottled water are exemplary pathways for microplastic entry into the human body (Issac & Kandasubramanian, 2021). Recent studies of microplastics in human stool, placenta (Ragusa *et al.*, 2021), and blood (Hwang, Choi, Han, Choi, & Hong, 2019) are examples of their presence in human. The accumulation of microplastic particles in human has potential health risks such as cytotoxicity, hypersensitivity, unwanted immune response, and acute response such as hemolysis (Hwang *et al*).

The Volta River Basin is in West Africa, and covers an estimated area of 400,000 km^2. The river basin stretches from latitude 5° 30' N in Ghana to 14° 30' N in Mali. The Volta Basin is shared by six West African countries (43 % in Burkina Faso, 42 % in Ghana, and 15 % in Togo, Benin, Cote d'Ivoire and Mali). In Ghana, the Volta Lake is a major freshwater ecosystem, and forms part of the Volta Basin system in the country. The Volta Lake reservoir has a surface area of about 8,500 km^2, an average depth of about 18.8 m and a shoreline of about 5,500 km (Barry, Obuobie, Andreini, Andah, & Pluquet, 2005). The Volta Lake, at its formative years, was divided into eight segments called strata to facilitate hydrological and limnological studies (Asmah, Karikari, Abban & Ofori, 2014; Béné, 2007). The climate of the basin is tropical continental or savanna type, with a single rainy season in the northern sector. However, there are two rainy seasons in the southern portion of the Lake extending from May to October, followed by a prolonged dry season. The annual rainfall ranges between 1000 mm and 1150 mm (Asmah *et al.*). The primary purpose of the Lake is for the production of hydroelectricity (Barry *et al.*). The Lake also serves as a source of water for domestic use, agriculture purpose, transport of goods and services, inland fishing, and commercial aquaculture production. Lake Volta hosts about 143 fish species and provides about 90 % of the total inland fish catch (Rurangwa, Agyakwah, Boon, & Bolman, 2015). Moreso, 80 % of the total aquaculture production in Ghana is from the Lake (Asmah *et al.*, 2014; Ministry of Fisheries and Aquaculture Development, 2012).

Aquaculture was introduced in the early 1950s in the Northern part of Ghana. Currently, it is practiced throughout the country, but is more concentrated in the central and southern parts. The main species of fish farmed in Ghana are: Nile Tilapia (*Oreochromis niloticus*) and African Catfish (*Clarias gariepinus*) (Odei, 2015). Until a decade ago, the culture was largely undertaken in earthen ponds, mainly at the subsistence level involving the use of rudimentary tools (Asmah *et al.*, 2014). This mostly consisted of land-based production units (earthen ponds) where fish farmers practiced extensive aquaculture production system. In such a system, fish is fed with low/poor quality feed, with a resultant low fish production (Odei, 2015). The need to increase fish production led to the introduction of the cage culture system. A fish cage is a confining facility with floats, frames, net etc. that can be mounted in an open waterbody (Cardia & Lovatelli, 2015). The fish cage system provides the best platform for aquaculture to be practiced on a much larger scale in Ghana (Odei, 2015). This provides the best option in bridging the gap between the demand for food (fish), and its availability in the country (Odei, 2015). Cage culture is believed to have the capacity to contribute considerably to the total fish production and food security in Ghana by providing employment, income, revenue and livelihood to the public, government and the surrounding communities (Karikari, Asmah, Ofori, Agbo, & Amisah, 2016).

In 1998, the first commercial aquaculture farm established in the Volta Lake introduced the cage aquaculture system in Ghana (Asmah *et al.*, 2014). A vigorous campaign and promotion by government agencies over the years has enhanced commercial interests of both foreign and local investors in the sector (Asmah *et al.*, 2014). Thus, currently, over 60 cage farms accounting for over 2, 278 cages are operating on the Volta Lake and mostly in Asuogyaman District in Eastern Region, with most small-scale cage farms clustered between Akosombo Dam and Kpong Dam (Rurangwa *et al.*, 2015). Clusters of small and medium-sized enterprise (SME) cage farms are also developing in areas such as Kpeve in South Dayi District of Volta Region, Sedom and Asikuma in Asuogyaman District and Akrusu in Upper Manya Krobo District of Eastern Region (Rurangwa *et al.*, 2015), Sogakope in South Tongu District in the Lower

Volta Basin (Ministry of Finance, 2021). Furthermore, Ghana's aquaculture grew the fastest, at an annual rate of 28 % from 2006 to 2019 due to cage farming from the Volta Lake. In 2018, with 76,600 mt of farmed fish produced (Ragasa *et al.*, 2022), 90 % of fish produced was obtained from cages (Amenyogbe *et al.*, 2018). In Ghana, notable waterbodies such as River Pra and River Ankobra that can equally be used for aquaculture production are facing challenges, ranging from pollution due to mining, to the reduction of water levels that compromise fisheries. According to Karikari *et al*. (2016), unlike Rivers Pra and Ankobra, the Volta Lake and Volta River are suitable for culturing tilapia as it has an exceptionally suitable and constant water quality, and a consistent all year-round warm temperature with low to no pollution (Karikari *et al.*, 2016). The water quality of the Lake is also generally safe and within the recommended limit for freshwater (Ghana Statistical Service, 2014; Olalekan *et al.*, 2015).

Fish farming has been promoted in Ghana to increase fish supply, and satisfy the national demand for fish as domestic fish landings meet 59 % of the demand. It is a chosen alternative to fish imports (The Ministerial Conference on Fisheries Cooperation among African States bordering the Atlantic Ocean, 2012). Fish is the preferred source of animal protein, and a central part of Ghanaian cuisine, providing approximately 60 % of animal protein consumed in the Ghanaian diet. The annual per-capita fish consumption of Ghana was estimated to be 25 kg in 2018 (United States Department of Agriculture, 2022).

Statement of the Problem

MPs are easily ingested by aquatic organisms from different trophic levels due to their size range (1-5 mm). Thus, they can enter the food chain, accumulate at higher trophic levels, and pose a potential risk to aquatic organisms and human health (Elizalde-Velázquez & Gómez-Oliván, 2021). The ingestion and concentration of microplastics in an organism are influenced by the availability and concentration of MPs in the environment. Microplastic concentrations in waterbodies can be influenced through discharge from wastewater plants for treatment, weathering and degradation of plastic waste in the bodies of water, and soil erosion, or runoffs from terrestrial inputs (Acquah *et al.*, 2021). On the Volta Lake, cages are the dominant culture fish production system. Most of these cages have components such as galvanised pipe frames, nets, ropes, lines and buoys/floats, drums, etc. made from plastics. (The polymers constituting the various plastic components are- Barrel/float-High density polyethylene (HDPE), Nets- Polyethylene, nylon, polyester, Polyethylene terephthalate, polypropylene, Ropes- Polypropylene, polyethylene, low-density polyethylene (LDPE), High density polyethylene (HDPE) (Lusher *et al*.,2017). These plastics can degrade or by biofouling, break-off with a resultant release of secondary microplastics. This might increase the occurrence and abundance of microplastics within and around the cages both vertically and horizontally in the water column and eventually settle in the sediment and act as long-term sinks for MPs.

Purpose of the study

The aim or primary objective of this study is to investigate the spatiotemporal distribution of MPs pollution in the Lower Volta Basin in Ghana, in cultured and wild fish species, and to assess the potential impact on fish consumers.

Research Objectives

This research seeks to:

i. Extract and quantify MPs ingested by cultured and wild fishes
ii. Extract and quantify MPs in fish feed used at sampled farms.
iii. Determine human exposure risk on consumption of fish contaminated with MPs.

Research Questions

1. Does aquaculture installations and inputs contribute to the different types and concentrations of MPs in the Lower Volta Basin?
2. What level of MP concentrations and types do cultured and wild fish species in the Lower Volta Basin ingest?
3. Does MPs in fish from the Lower Volta Basin induce exposure risk to human consumers?

Hypothesis

This study addresses the following hypotheses:

Null hypothesis

i. Aquaculture installations and inputs (cage net material, feed, etc) do not influence and contribute to the different types and concentration of MPs in the Volta Lake, and/or the Lower Volta Basin.

ii. Cultured and wild fish do not ingest different types and concentration of MPs in the Lower Volta Basin.

iii. Humans are not exposed to MPs on consumption of MP-contaminated fish.

Alternative hypothesis

i. Aquaculture installations and inputs (cage net material, feed, etc) influence and contribute to the different types and concentration of MPs in the Volta Lake, and/or the Lower Volta Basin.

ii. Cultured and wild fish ingest different types and concentration of MPs in the Lower Volta Basin.

iii. Humans are exposed to MPs on consumption of MP-contaminated fish.

Significance of the Study

This study investigates the presence and impact of microplastics on cultured fish in cages, and wild fish in the Lower Volta Basin, and the Volta Lake, and their health implications upon fish consumption by humans.

Although there is increasing interest in the study of MPs, most research and publications are focused on the marine environment; very few studies focus on freshwater ecosystems, globally. The first study on freshwater microplastics was reported in 2011, with an emphasis on Lake Huron and Los Angeles rivers in the United States (Zbyszewski & Corcoran, 2011). In Africa, including Ghana, there is insufficient data on the extent of microplastics in freshwaters (Acquah *et al.*, 2021). To date, only ten studies have been completed on MP studies in Ghana. Of these ten, seven (70%) were reported in the marine and three (30%) in freshwater environments. Studies focused on the marine environment were done in brackish ecosystems such as Mukwei Lagoon, Kpeshie Lagoon, Sakumono Lagoon, Mangrove Forest remnant to coastal waters of the Gulf of Guinea at James Town, Tema, Sekondi, Denu, amongst others. These studies were reported by scientists including Adika *et al*. (2020), Gbogbo, Takyi, Billah, & Ewool, (2020), Chico-ortiz, Mahu, Crane, Gordon, & Marchant, (2020), Faseyi, Miyittah, Yafetto, Sowunmi, & Lutterodt, (2022) and Pappoe *et al*. (2022). Samples taken comprised of fin fish, oysters, water, sediment, salt etc. Microplastic contamination was recorded across all samples taken; an indication of MPs contamination in all sourced ecosystems. Sizes of MPs recorded in the marine environment ranged from 1.2 µm to 5 mm. Microplastic types identified included fibres, fragments, foam and pellets (Adika *et al*; Chico-Ortiz *et al.*; Faseyi *et al*; Gbogbo *et al*; Pappoe *et al.*).

Furthermore, freshwater studies were reported by Blankson *et al*. (2022), Adu-Boahen *et al*. (2022) and Faseyi *et al*. (2022) on the Weija Dam, Densu Delta, Akora River, Pra and Anokobra estuaries (Adu-boahen, Dadson, Kwaku, & Mensah, 2022; Blankson, Tetteh, Oppong, & Id, 2022; Faseyi *et al.*, 2022). Microplastic contamination were recorded across all study samples of fin fish, water and sediment. Microplastic sizes stated ranged from 0.3-1.2 mm which were measured from MP shapes of fragments, sheets, fibres and beads. Microplastic identification in both study environments were done by visualization with microscope (fluorescent, dissecting, light etc.) and FTIR (for characterization of polymers, extensive work completed beyond the scope of this Master's thesis; results are not reported in this thesis). Misidentification, over-estimation of MPs abundance and failure to characterize MPs shapes and polymers were some of the limitations in previous studies that needs to be addressed.

Thus, the limited studies on MPs in Ghana, and most especially in the freshwater environment coupled with limitations mentioned above in previous studies justify the need for this study. Moreso, this study aims to provide information on the occurrence and spatiotemporal distribution of MPs in farmed and wild fish species, sediments and river water from freshwater ecosystem of the Lower Volta Basin in Ghana. A summary of the studies on MPs completed in Ghana, to date, has been compiled in Table 2.

Delimitation

All sampling sites were from the Lower Volta Basin with no consideration of sampling points from the Upper Volta Basin. The three sites sampled or chosen included a site above the Akosombo Dam (Asikuma Labolabo), another site below the Dam (Kpong), then a site close to the mouth of the Lower Volta Basin (Sogakope Agodormi) to observe possible differences in the spatiotemporal distribution of MPs at these sites. Furthermore, sampling points chosen were based on the presence/availability of aquaculture farms. Morphometric data on fishes sampled were taken, excluding the age of the fish.

Data on MPs shape/type, colour and size were analysed in all sample matrices. Characterization of MPs based on polymers were not included in this write-up.

Limitations

Analysis on fish from the cultured and wild sources were not species specific (For instance, wild species of *Oreochromis niloticus* were not obtained). The age group of the fish species were not considered, thus the data obtained will not highlight how MPs affect a particular species or age group.

For sampling, funds allocated for the project from which this data was collected, faced delays in disbursement, coupled with the constraint of a short project duration- limited to 12 months. As a consequence, the sampling frequency was restricted to one event at each site for each season.

Definition of Terms

Species specific: same fish species from the cultured and wild source.

Age specific: same age group or size of fish from the cultured and wild source.

Chapter Summary

This thesis has six (6) chapters. Chapter One (1) introduces the whole concept of the study, giving the background and stating the problem, purpose, objective and significance of the study. Chapter Two (2) reviews literature which pertains to the study including the methodology used. In Chapter Three (3), the study design, and methods employed are described. The study areas are clearly mapped out and statistical tools and application for data collection and analysis are explained. The results obtained in the study are presented in tables and figures with brief description in Chapter Four (4). Analysis and inference are drawn in Chapter 5 in a form of discussion. Conclusions and recommendations have been given in Chapter Six (6). Other sections presented in the thesis include the references and appendices.

CHAPTER TWO

LITERATURE REVIEW

Fish Production from Freshwater

Freshwater ecosystems (lakes and rivers) hold less than 0.02 % of all water on earth, and occupy about 6 % of the land surface area. Surface waters are constantly replenished and easily accessed and provides the needed water supply for the vast majority of humans on earth. In addition to providing the water necessary for crop growth, inland waters support freshwater fisheries, and provide about one-third of global annual fish catch (Schlesinger & Bernhardt, 2020). Positively, inland fisheries (both wild and aquaculture) have produced about 63.3 million tonnes of fish worldwide (as at 2018) (Food and Agriculture Organization, 2020). In Africa, the Nile River in Egypt (North Africa), Lake Chad Basin in Nigeria (West Africa), the Great lakes of the rift valley in Eastern and Southern Africa are part of notable freshwater ecosystems that have contributed to 2.1 million tonnes of inland fish production (Neiland, Chimatiro, Khalifa, Ladu, & Nyeko, 2005). As at 2014, Nigeria, Uganda, Ghana, Kenya and Zambia were among the top inland fishery producers. In Ghana for instance, more than 90 % of the inland freshwater fish, and 80 % of the total aquaculture production are obtained from the Volta Lake which is a dominant feature of the Volta Basin system (Béné, 2007). Tilapia (*Oreochromis niloticus*) and catfish (*Clarias spp.*) are the dominant species cultured. Cages dominate the culture production facilities on the Volta Lake (Asmah *et al.*, 2014). The composition of the commercial catch/fish landings comprise: *Chrysichthys spp.*, *Synodontis spp.*, *Labeo spp.*, *Mormyrid spp.*, *Heterotis spp.*, *Clarias spp.*, Schilbeids, *Odaxothrissa mento*, *Bragrus spp.*, *Citharinus spp.*, *Alestes spp.*, *Brycinus spp.*, *Distichodus spp.*, *Gymnarchus spp.*, *Hydrocynus spp.*, and *Lates niloticus* (Béné, 2007; Ministry of Food and Agriculture, 2003). The main types and number of fishing gears used include gill nets, line fishing, traps, cast nets, nifa nifa, beach seines, winch nets, atigya etc (Béné, 2007). Fishery is solely artisanal with about 17,500 canoes actively fishing on the Lake operating from about 2,000 fishing villages. It is estimated that a total of 300,000 people depend on the lake for their livelihood- of which 80,000 are fishers, 20,000 fish processors and traders, and the rest farmers, livestock farmers, petty traders, and food processors (Béné, 2007). Inland capture fisheries and aquaculture contribute 30 % to the total domestic fish production (Asiedu *et al.*, 2017). An estimated 80 % of total domestic fish production is consumed locally, and it constitutes about 60 % of total animal protein intake (Asiedu, Berchie, Nunoo, & Iddrisu, 2017). The contribution of fisheries to livelihoods and food security increased from 2.2 to 2.4 million people between 2011 and 2015. The sector generates US$ 1 billion revenue yearly (Ministry of Fisheries and Aquaculture Development, 2015). Foreign exchange earnings from fisheries increased from US$ 165.7 million in 2010 to US$ 309.7 million in 2015, with a corresponding increase in the overall fish production by a volume of 9.3 % between 2010 and 2015. The Ghanaian fisheries contributed about 1.5 % to the GDP annually, at a GDP growth rate of 5 % (Ghana Statistical Service, 2015).

Freshwater Resource in Ghana

Ghana is well endowed with freshwater resources. Apart from the Volta River basin which constitutes 70 % of the country's drainage system, other river basins include the Densu River Basin, Ankobra basin, Pra Basin, Tano Basin and White Volta Basin (Institute of Statistical Social and Economic Research, 2020). These freshwater resources are mainly used for domestic consumption, irrigation, livestock watering and fish farming. The growing population and constant exploitation of freshwater resources to meet the above-mentioned needs of humans have threatened/endangered the resource (Yeleliere *et al.*, 2018). The issue of climate change coupled with environmental pollution from waste (both municipal and industrial waste), leaching of toxic chemicals from fertilizers and pesticides used in agriculture, has further deepened the woes in the potential use of this freshwater resource (Yeleliere, Cobbina, & Duwiejuah, 2018). In Ghana, the recent increase in galamsey (illegal mining) has led to the deterioration of the quality of most of the freshwater resources in the country (Eduful *et al.*, 2020), with some of the most affected including Offin, Pra, Ankobra, Birim, and Tano Rivers (Attiogbe & Nkansah, 2017). In spite of the challenges facing the freshwater ecosystem in the country, the Volta Lake/River remains one of the resources which has been least impacted by anthropogenic activities, and has a constant quality all year round to support capture fisheries, as well as aquaculture (Karikari *et al.*, 2016).

Sources of Microplastics in Freshwater Bodies

Freshwater bodies are highly managed for generation of electricity, recreation, supply of potable water, and transport of goods and services (Schlesinger & Bernhardt, 2020). In the wake of all its usefulness, inland and freshwater ecosystems face a lot of threats. Some of these threats include overexploitation, flow modifications, destruction or degradation of habitats, invasion by exotic species, and water pollution (including microplastics pollution) (Dudgeon *et al.*, 2015). Microplastic pollution has become an issue of interest due to mass production, mass utilization of plastics and improper plastic waste disposal. This has led to the accumulation of plastics in natural habitats, coupled with adverse impacts on biota and the economy (Lusher *et al.*, 2017). Moreover, MPs are ingested throughout the food web more readily than larger particles (Wagner *et al.*, 2014). Globally, a total of 8 million tonnes of plastic waste leaks into the ocean each year. In Ghana, out of 1.1 million tons of plastic waste generated per year, only 5 % is collected for recycling (World Economic Forum, 2021), with the rest mostly finding their way into waterbodies. These plastics are durable, and do not biodegrade (Acquah et al., 2021).

Apart from the contribution of plastic waste, activities of fisheries and aquaculture contribute microplastics to waterbodies. In fisheries, plastic materials are used in boat construction (including painting and anti-fouling coats), boat maintenance, fishing gears (nets, trawls, dredges, traps, floats, lures, hook and lines), fish hold insulation and fish crates (Fisheries and Aquaculture Organization, 2016). Nets and floats are made from a range of plastics polymers, including polypropylene (PP), polyethylene (PE), polyvinyl chloride (PVC), polystyrene (PS), and polyamide (PA) (Lusher *et al.*, 2017). Aquaculture done in open waters such as rivers, lakes, estuaries and other coastal waters make use of facilities such as cages and pens which comprise of nets stretched over a framework structure. The nets, framework, and other equipment such as floats, ropes, lines, buoys, and barrels are primarily made of plastic material. As this equipment are continuously used, they are exposed to direct UV light, wave action, abrasion and temperature changes. These factors can contribute to embrittlement and fragmentation resulting in the loss of large plastics and the formation of microplastics.

Another source of microplastics in the fisheries and aquaculture sector in open waters has to do with abandoned, lost or otherwise discarded fishing gears (ALDFG). These gears include gillnets, trawls, handlines and longlines used in fishing. Loss of these fishing gears can be a result of enforcement on fishers to abandon gears (e.g. illegal fishing or illegal gears), operational pressure (e.g. use of too much gear in restricted time periods) and environmental conditions (e.g. weather, waterbody bed irregularities), lack of/inaccessible/expensive onshore gear and waste disposal facilities (Gilman, Chopin, Suuronen, & Kucmlangan, 2016; Macfadyen & Allison, 2009). In aquaculture, polyvinyl chloride (PVC) tubes, net caps, plastic bands, zip ties, ropes, and floats are lost due to wear and tear of anchor ropes, storms, and accidents or conflicts with other water users. These lost items end up in the water column or settle on the waterbed or are washed on-shore where they continue to breakdown leading to the formation of microplastics which could have further impact on the environment. The continuous removal of biofouling organisms from aquaculture facilities, as they are being cleaned, could release net and rope fibres in the form of microplastics to the environment (Gilman *et al.*, 2016).

Occurrence and Spatiotemporal Distribution of Microplastics in Freshwater

As mentioned earlier, most research on MPs focus on the marine environment; however, few studies from freshwater environments show that freshwater or inland water face similar issues with MP contamination as the marine environment. Due to the connection/interlinkages between the water system, freshwater bodies such as lakes, rivers and streams act as transport pathways for debris into the ocean (Lebreton *et al.*, 2017). In freshwater environments of lakes and rivers, studies also report highly heterogeneous concentrations of MPs comparable to those reported for the marine environment (Wagner & Lambert, 2018). Literature reviewed showed that studies completed in inland waters in Asia, America, Europe, Africa reported the occurrence of MPs such as fragments, pellets, films, foam fibres etc. For instance, a study by Rodrigues *et al*. (2018) in Portugal, classified isolated MPs sampled from the Antuã River in fragments, pellets, films, foam and fibres. They used physical characteristics such as colour of particles- white, transparent, and black (Rodrigues *et al.*, 2018). Likewise, Tang, Gao, Gao, & Zou, (2021) also grouped MPs sampled from the Songhua River in China into fibres, films, pellets, foams and debris (Tang *et al.*). In Ghana (Africa), however, a study done on the Akora River by Adu-Boahen *et al.* (2022) did not classify or categorize the microplastics found. The authors reported the occurrence and abundance per volume of water of MPs at all the five (5) sites of the river sampled. In their study, site 1 recorded 67 items of microplastics per 2.8 m3, site 2 recorded 79 items per 6.3 m3, and site 3 recorded 73 microplastics per 3.7 m3. Sites 4 and 5 recorded 41 and 26 microplastics per 2.8 m3 and 1.8 m3, respectively (Adu-Boahen *et al.*, 2022). Similarly, in studies of Blankson *et al.* (2022) on the Weija Dam and Densu Delta, the average MP abundance reported in the two systems were 1.583 ± 0.167 items/60 mL and 4.16 ± 0.342 items/60mL, respectively.

They equally did not report the MP type or shape but reported on the size of MPs- Weija Dam: 1.21 ± 0.26 mm and Densu Delta: 1.01 ± 0.11 mm.

Although different factors affect the spatial and temporal distribution of microplastics in freshwater, most factors reported from literature include anthropogenic activities- high industrialization and urbanization, wastewater treatment plants (WWTP), environmental conditions of wind, rainfall (run-offs), hydrodynamic conditions and methodological approaches. In the study of the Antuã River, the sites were selected based on their high and low population density, urban and industrial area, as well as the proximity to wastewater treatment plants (WWTP). The results showed that high levels of MP contamination and variability (in terms of abundance) were related to conditions at the sampling site- proximity of the sampling sites to sources of MPs such as industry, urbanization and waste water treatment plant. This increases the likelihood of MP appearance in all sites, related to the flow velocity, specific characteristics of plastics, biofouling and adsorption of substances which might influence their physical and temporal detection. Additionally, Tang *et al*. (2021) reported that with reference to the 39 sampling sites selected along the Songhua River, densely populated urban and tourist areas and tourist had high microplastic abundance due to human interferences from fishing, swimming, and leisure activities. The abundance of MPs in different sampling sections were significantly different ($p < 0.01$) and increased with the intensity of human interferences. The study concluded that human activities were important sources of MPs along the river. Besides human activities, accumulation of MPs were observed along the flow of the river, thus influencing the abundance and spatial distribution of MPs at these sampling sites (Tang *et al.*, 2021). Furthermore, Adu-Boahen *et al.* (2022) observed that three out of the five sampling sites on Akora River had higher abundance of microplastics due to direct human interaction with these sites. They also identified that gutters/drains around the river that serve as wastewater channels feed the river with the microplastics, and serve as the main source of pollution.

Microplastics in Freshwater Sediment

Microplastics with a density higher than 1.0 g/cm^3 sink, and get deposited in sediments, while low-density debris float on the surface of water and/or within the water column. Studies have suggested that the accumulation of biofilms, the adsorption, and accumulation of pollutants lead to an increase in the density of polymer debris, which is the main reason for the appearance of microplastics in sediments (Yang, Zhang, Kang, Wang, & Wu, 2021). A review completed by Yang *et al.* on the methods, occurrence and sources of microplastics in freshwater sediments reported that MPs contamination of freshwater sediment is on a global scale and occurs in different freshwater bodies including rivers, lakes and reservoirs. The review categorically reported on MPs in sediments on river banks, river bottom, lake shore, and lake bottom as well as reservoirs. The average abundance of freshwater sediment microplastics vary widely among different study areas and matrices. As observed within the water column, the abundance of microplastics in densely populated areas was higher than that in sparsely populated areas (Yang *et al.*, 2021).

Peng, Xu, Zhu, Bai, & Li, (2018) reported the average microplastic concentration of a densely populated Shanghai urban district area to be 802 ±

594 items/kg of sediment. On the other hand, Zhang *et al*. (2021) reported MP abundance in sediments at 10 sites along the lower reaches of Qin River, a less populated catchment area (Peng *et al.*, 2018). Microplastic concentration ranged between 0 and 97 items/kg (Zhang *et al.*, 2021). The highest reported value in riverbank sediment was recorded in Rhine-Main River, Germany- MP concentrations of 260 ± 10 to 11,070 ± 600 items/kg were identified (Mani *et al.*, 2019). In Africa, Nel, Dalu, and Wasserman, (2018) found concentration of microplastics in river bank sediment of Bloukrans River in South Africa to be 660 items/kg. Similar to riverbank sediments, the presence of microplastics has been recorded in river bottom sediment worldwide (Yang *et al.*, 2021). China is projected as a hotspot of plastic pollution (Jambeck *et al.*, 2015; Xu, Xiang, & Hac, 2020). Wang, Liu, Chen, and Xia, (2019) recorded an extremely high concentration of microplastics (32,947 ± 15,342 items/kg) in the bottom sediment of the Wen-Rui Tang River (Wang *et al.*). Rodrigues *et al.* (2018) detected MPs in the bottom sediment of the Antuã River in Portugal to be 18 to 629 items/kg; the study emphasized the importance of rivers as a potential transportation system for microplastics (Rodrigues *et al.*, 2018). A study by Blankson *et al.* (2022) on the Densu River and the Weija Dam in Ghana reported concentrations of 4.00 ± 0.82 per 10 g and 3.75 ± 1.71 per 10 g of MPs, respectively. The study stated that although the Densu Delta is a lotic system and the Weija Dam a lentic system, the mean number of microplastics in sediments of the two systems were statistically similar. Thus, these systems were not affected by differences in their hydrology (Blankson *et al.*, 2022).

Lakes are relatively closed, natural waterbodies that can store water from precipitation, surface runoff, and groundwater (Yang *et al.*, 2021). Plastic garbage generated in lake catchment can be transported to the lake and accumulate there (Su *et al.*, 2016; Yang *et al.*, 2021; Zhang *et al.*, 2019). A high abundance of 12,000-200,000 items/kg of MPs was recorded in the lake shore sediment in urban recipient in Norway (Haave, Lorenz, Primpke, & Gerdts, 2019). The major source of MPs was considered to be sewage outlets (Zhang *et al.*, 2021). Zhang *et al.* (2016) also found a high concentration of microplastics (4–1219 items/m2) in the remote area of the Tibet Plateau in China to be mainly derived from mismanaged plastic wastes (Zhang *et al.*, 2016). The concentration of microplastics in lake sediments was concentrated in the range of 11–2175 items/kg (Zhang *et al.*, 2021). In line with this, a study by Yuan *et al.* (2019) obtained MP concentration in the range of 54–506 items/kg in Poyang Lake in China (Zhang *et al.*, 2021). A report also showed that a total of 4635 microplastic debris were found in Lake Ontario in Canada (Vermaire, Pomeroy, Herczegh, Haggart, & Murphy, 2017). Bordos *et al.* (2019) found relatively low concentrations of MPs (0.46-1.62 items/kg) in the Central Eastern European inland lakes; this was the first study completed on fish ponds whose results indicated that fish ponds act as deposit areas for microplastics.

Occurrence of Microplastics in Fish

Fish is an important biological element of freshwater ecosystems with significant economic and nutritional value worldwide (Pinheiro, Oliveira, & Vieira, 2017). Fish can ingest MPs in two ways: directly (when consuming prey or attacking items resembling prey) or indirectly (by ingesting prey which contains MPs). Recent studies suggest that fish feeding type and behaviour is related to the MP content in the fish (Kuśmierek & Popiołek, 2020). A study by

Zhang *et al*. (2021) on MPs in freshwater and wild fishes from Lijiang River in Guangxi, Southwest China reported that among the 84 individual fishes belonging to 4 species of *Cyprinus carpio, Pelteobagrus fulvidraco, Mystus macropterus*, and *Pelteobagrus vachelli*, MPs were identified in 68 individual fishes (81.0 %) with an average abundance of 0.6 ± 0.6 items/individual. Moreover, they reported that *Mystus macropterus* which ingests benthic aquatic insects and larvae, snails, as well as little fishes and shrimps had MP abundance (0.9 ± 0.3 items/g) that was significantly higher than those in other three fish species which are surface feeders ($p < 0.05$). Thus, they ascribed their observation to the different feeding and living habits of different fish species, resulting in distinct distribution of MPs in the fishes. In their study, MPs in the morphotypes of flakes and fibres were dominant in both water and fish samples with PET MPs being dominant in the fishes (Zhang *et al.*, 2021).

A study by Garcia, Suárez, Li, and Rotchell, (2021), which compared microplastic contamination in freshwater fish from natural and farmed sources reportedly found no significant differences in microplastics found in tissues of *Oreochromis niloticus* from aquaculture farms in the Huila region, and natural source fish (*Prochilodus magdalenae* and *Pimelodus grosskopfii*) from the Betania Dam, the Magdalena River in Columbia. However, a higher number of fishes sampled from natural sources presented MPs in their tissues (75 %) compared to farmed fish (44 %). The authors attributed this to the difference in habitat, as the natural sourced fish were characterized as having benthopelagic habits while the farmed source as epipelagic. Secondly, the Magdalena River from which the wild fish were sourced passes through a city (Neiva) characterized by dense human settlement and untreated wastewater discharge;

these are considered as potential sources of MPs into the River. Furthermore, fragments were the main type of MPs observed in gut, gill, and flesh of both sources of fish. Polyethylene terephthalate, PS, and PE were the main polymers from the chemical composition of MPs isolated from the tissue (Garcia *et al.*, 2021).

A study by Ilechukwu *et al*. (2021) on *Chrysichthys nigrodigitatus* from Calabar River in Niger Delta, Nigeria, reported that microplastics were in the guts of twenty-five (25) samples (56 %) out of 45 samples examined. With the MPs observed, 87.93 % were fragments, 10.92 % were fibres, and 1.15 % were pellets (Ilechukwu, Ndukwe, Ehigiator, Ezeh, & Asogwa, 2021). They reported that the dominance of fragments came from solid domestic and municipal waste that are emptied into the River.

A review by Uzomah, Lundebye, Kjellevold, Chuku, and Stephen, (2021) reported that there was limited data available regarding microplastics in fish from Nigeria; however, a recent study was published by Adeogun *et al.* (2020) on fishes sampled from Eleyele Lake in Ibadan (Oyo State) in southwestern Nigeria. In the study, a total of 109 fish sampled belonging to eight species *(Coptodon zillii, Oreochromis niloticus, Sarotheron melanotheron, Chrysichthys nigrodigitatus, Lates niloticus, Parachanna obscura, Hemichromis fasciatus and Hepsetus odoe*) were analyzed from different habitats and trophic levels. From their findings, all of the species except *Hemichromis fasciatus* had microplastics in their stomachs. The highest occurrence of microplastics was reported in benthopelagic fish species, *O. niloticus* (34 %*)*, then, *C. zillii* (32 %), and *S. melanotheron* (13 %). The lowest occurrence was found in *P. obscura*, *L. niloticus*, and *H. odoe*- (5 %) (Uzomah *et al.*2021). They reported that the different percentages of MPs in the various species was influenced by ecological variables such as habitat, feeding mode, and trophic levels (Uzomah *et al.*, 2021).

Another study by Dada and Bello (2023) on microplastics in carnivorous fish species, water and sediments of a coastal urban lagoon in Nigeria reported that MPs were found in water, sediment, and three carnivorous fish species (*Hepsetus odoe*, *Chrysichthys nigrodigitatus*, and *Lachnolaimus maximus*). This raised concerns about the health of the Nigerian Lagos Lagoon ecosystem and the human food chain. They also reported that fibres were more concentrated in the water, while fragments predominated the sediment matrix. The source of MP pollution in the water were plastic debris from recreational, industrial, and domestic wastes (Dada & Bello, 2023).

Another study as mentioned earlier by Adu-Boahen *et al*. (2022) in mapping ecological impacts of microplastics on freshwater habitat in the Central Region of Ghana: a case study of River Akora involved the analyses of 164 individual fishes harvested from Akora River- *Oreochromis niloticus*, *Oreochromis aureus*, *Oreochromis mossambicus*, *Sarotherodon melanotheron* and *Clarias anguillaris*. They reported that out of 164 fishes sampled, 21 had ingested microplastics with the average count of ingested microplastics being 30 pieces per fish. This confirmed the presence of MPs in the river, with potential bioaccumulation across the biotic food chain. The study did not categorize the MPs observed into shape/type nor establish the polymer types of MPs identified (Adu-Boahen *et al.*, 2022).

Occurrence of Microplastics in Fish Feed/Meal

The presence of microplastics (MPs) in a broad range of wild and cultured fish species has been well-documented, but transfer mechanisms by which cultured organisms are contaminated with MPs is poorly understood (Karbalaei *et al.*, 2019). For cultured species, one route of MP contamination can be through the fish feed/meal. Fish meal (FM) is an industrial product mainly obtained from whole wild-caught fish that is used as a high protein feed stuff component in aquaculture and intensive animal farming (Vázquez-rowe, Ita-nagy, & Kahhat, 2021). Only a few studies on MPs in fish meal have been published- the majority of which are reported in Asia, Europe, and America. There are no published MP-contaminated feed studies in Africa, and Ghana. A study by Karbalaei *et al*. (2019) reported that from three brands of Malaysian commercial fish meal analysed, fragments were the dominant form of MPs (78.2 %), followed by filaments (13.4 %), and films (8.4 %). The study demonstrated that cultured organisms could be exposed to high levels of MPs via MP-contaminated fish/shellfish used in fish meal production (Karbalaei *et al.*, 2019). In another study by Gündogdu, Eroldogan, Evliyaoglu, Turchini, Wu, (2021), reports on sampled fish meal from countries such as Antarctica, Chile, China, Denmark, India, Morocco, Mauritania, Norway, Peru, South Africa, South Korea and Turkey showed that there was a total of 52.6 % MP fragment type; 38.7 % were of fibre/filament type, 8.5 % film type, and 0.2 % were of foam type. In terms of quantity, the mean number of plastics isolated from fish meal per country ranged between 0 and 337.5 items kg^{-1}. The highest content of plastics was found in samples obtained from China (337.5 ± 34.5 number of items kg^{-1}). The lowest content of plastic was recorded in samples supplied from Norway (33.3 ± 6.7 number of items kg^{-1}) and South Korea (33.3 ± 6.7 number of items kg^{-1}). Additionally, no plastic was found in krill meal sample obtained from Antarctica (Gündoğdu *et al.*, 2021).

From studies of Hanachi, Karbalaei, Walker, Cole and Hosseini, (2019) focused on commercial fish meal and cultured common carp (Cyprinus carpio) from factories in Southern Iran, MP fragments were the most predominant morphology (67 %). This was followed by films (19 %), pellets (8 %), and fibres (6 %). They further reported that the most abundant plastic polymer in fish meals were polypropylene (PP) (45 %), followed by polystyrene (PS) (24 %), polyethylene (PE, 19 %), polyethylene terephthalate (PET, 8 %), and rayon (4 %). The study highlights that marine-derived fish meal may be a source of MPs which can be transferred to cultured fish, posing a concern for aquaculture. The study also pointed out the fact that *Cyprinus carpio* can be used as an effective bioindicator to reveal the presence and transfer of MPs from the marine environment into humans within the food chain (Hanachi *et al.*). The few studies reviewed by Karbalaei *et al.* (2019), Hanachi *et al*. and Gündogdu *et al*. (2021) mentioned that cultured organisms could be exposed to high levels of MPs via MP-contaminated fish/shellfish used in fish meal production. This is because fish meal is mainly produced with small pelagic species, by-catches, excess allowable catch quotas trimmings, and fish processing wastes (Mahamud *et al.*, 2022).

As fish are small, exposure to MPs result in endogenous contamination, as MPs are not expelled from the gastrointestinal tract, and exogenous where MPs are possibly present as marine litter (Castelvetro *et al.*, 2021). Most of the literature reviewed recommended that fish meal replacement with other sources of protein including meat meals and plant-based meals may be considered to reduce the level of exposure (Gündoğdu *et al.,* 2021; Hanachi *et al.,* 2019; Karbalaei *et al.,* 2019).

Human Exposure Risk Assessment

The presence of accumulated microplastics in fish may not only reflect contamination in the aquatic ecosystem, but also mirror the potential dangers to human health posed by the ingestion of seafood that contains microplastics (Pappoe et al., 2022) . As fish is used in human food table across the world, they constitute a long-term exposure route for all humans and raise the concern about potential public health risk. Accumulation and distribution of MPs by commercially important aquatic organisms is expected to lead to greater exposure risk for human populations with possible adverse effects over time (Makhdoumi, Hossini & Pirsaheb, 2023). The extent of human exposure to microplastics is computed, by the use of Intake index. This is done by using the mean number of MPs per kilograms per day in fish muscle to estimate the daily intake (EDI).

A review by Makhdoumi *et al*; (2023) reported that MPs were found in 56.5% of the commercial fish samples analysed across the world. They reported that the EDI of adults and children that consume such contaminated fish to be around 232,260 to 8,864,100 P/kg/bw/year and 25,280 to 964,800 P/kg/bw/year respectively.

Another review was conducted by Cox et al., (2019) to identify studies that determined the concentration of microplastic particles within food and beverages consumed by Americans. They stated that twenty-six studies were identified specifically investigating the consumption of fish, shellfish, added sugars, salts, alcohol, water, and air. From these twenty-six studies, they indicated that American adults and children consuming the recommended or average amounts of the items that have been analyzed for MPs are exposed to between 81000 and 123000 MPs per year.

A study by Pappoe et al., (2022) in Ghana reported annual consumption of microplastics in fishes they investigated an follows -*D. rhonchus* to be 1307 to 3920 microplastic particles per year, while annual intake by Ghanaian population through consumption of *S. maderensis* could range from 3063 to 15,313 particles per year. The consumption of *P. prayensis* and *P. bellottii* could expose the population from 4941 to 29,647, and 3111 to 15,556 microplastic particles per year, respectively.

A different study by Addo et al.,(2022) on the occurrence of microplastics in wild oysters (Crassostrea tulipa) from the Gulf of Guinea in Ghana reported that based on the average MP item per gram tissue (1.0 g/tissue), the estimated mean human intake of microplastics for the general population consuming 50 g of the assessed species was 50 MP items/week or 2,600 MP items/year, corresponding to 1.0 MP item/g/week and 52 MP item/g/year.

Some studies stated that the toxic effect of microplastics on human health is not well understood, but could be manifested by interfering with human body functions such as digestive system, cardiovascular system, endocrine system and the nervous system. Therefore, further studies are needed to highlight the toxicity of plastic exposure to human health (Addo et al., 2022).

CHAPTER THREE

RESEARCH METHODS

This chapter describes procedures and techniques used to collect and analyse data during the study. It touches on the study area, sampling procedures, laboratory analyses, data processing and analytical tools applied.

Study Area

The Lower Volta Basin

The study area lies between longitude 0° 00’ and 0° 40’ E and Latitude 6° 00´ N and 6° 20´ N on the Lower Volta Basin (Figure 1). The entire Volta Basin covers a surface area of 409,000 km^2 with a mean depth of 18.8 m (Barry, Obuobie, Andreini, Andah, & Pluquet, 2005). The Volta Basin drains about 70 % of the total land area of Ghana, with the portion in Ghana representing 42 % of the total basin area (Asmah, Karikari, Abban & Ofori, 2014). The Lower Volta Basin in Ghana is within the Southern Savannah climatic zone, and it experiences two rainy seasons from March to November with peaks in May/June and October (Logah, Amisigo, Obuobie, & Kankam-Yeboah, 2017). Agriculture is the major land use activity in the basin, with most of the inhabitants of the basin being farmers engaged in both cultivation of crops and livestock rearing. The remaining areas are characterized by extensive livestock grazing (Asmah *et al.*). Recently, however, cage fish farming activities have been ongoing on the Lower Volta Basin. This is due to the quality of the water being exceptionally suitable for tilapia culture and having a consistent year-round warm temperature (Karikari, Asmah, Agbo, & Amisah, 2016). The study was conducted in three communities- Asikuma Labolabo, and Kpong in the

Eastern Region and Sogakope Agordomi in the Volta Region. Asikuma Labolabo is a small community/settlement along the bank of the Lower Volta Basin. The settlement is located above the Akosombo Dams. Kpong on the other hand, is below the Akosombo Dam, and noted for its fish landing site. Sogakope Agodormi is a small community close to the mouth of the Lower Volta Basin.

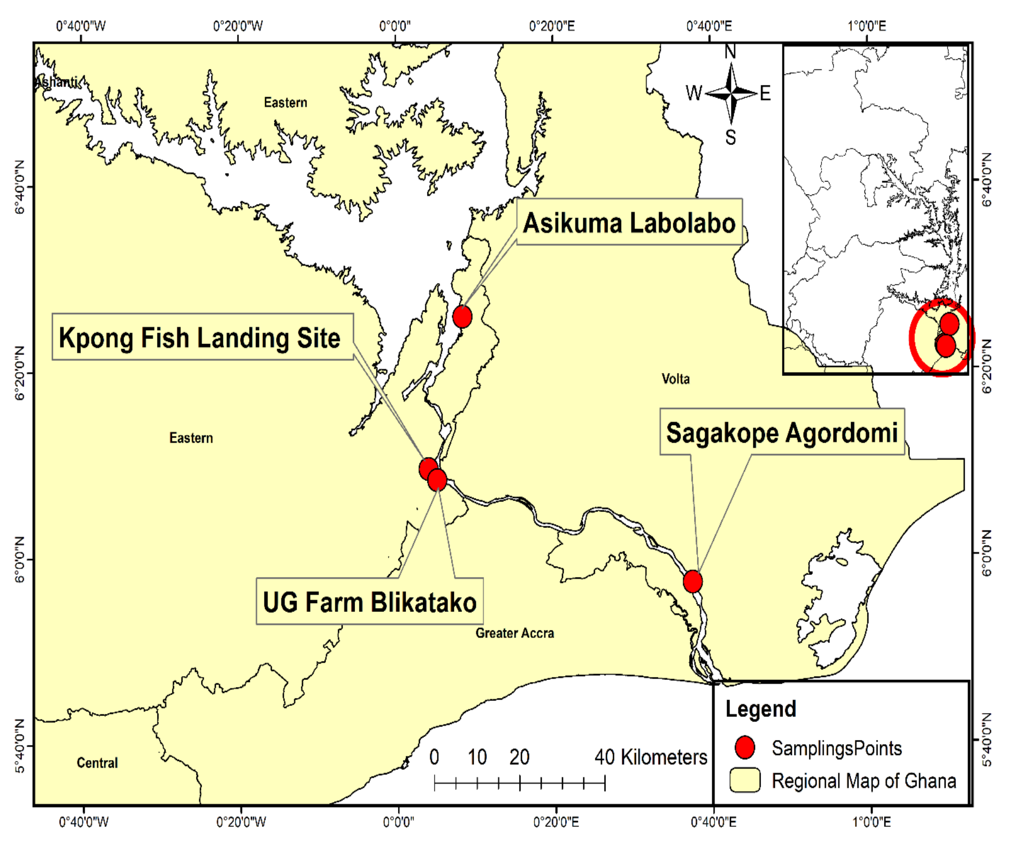

Figure 1: Map showing sampling sites on the Lower Volta Basin

Sampling Fish

Fish samples were purchased from cage fish farms for cultured species, and wild fish samples were bought from landing sites of fishermen at Agordomi, Kpong and Asikuma. Samples were taken in early April and August of 2022 for the dry and wet seasons, respectively. At each farm, > 20 kg of fish was bought, and these constituted a total of about 330 individual fishes for both seasons. Nile Tilapia (*Oreochromis niloticus-Appendix 5,6*) was the only cultured species obtained. The wild species were purchased from fishermen at random. Fish species were selected or bought based on availability. The variation in species bought is expected to provide information on diversification in MPs uptake by species. A total of 86 individual wild fishes were obtained for both seasons. However, the total fish constituted of these species: *Chrysichthys nigrodigitatus* (26 individuals) (Appendix 6), *Chrysichthys auratus* (5 individuals) (Appendix 6), *Tilapia guineensis* (13 individuals) (Appendix 7), *Tilapia zillii* (6 individuals), *Mormyrus spp*. (6 individuals) (Appendix 7), *Synodontis vellifer* (4 individuals) (Appendix 6), *Synodontis schall* (3 individuals Appendix 6), *Parachanna obscura* (3 individuals), *Auchenoglanis occidentalis* (3 individuals), *Brycinus brevis* (3 individuals) (Appendix 8), and *Malapterurus electricus* (2 individuals).

Monofilament gillnet and basket traps are the predominant fishing gear used by the fishermen in all three communities. The samples were transported on ice in an ice chest to the Water Research Institute (WRI) fisheries laboratory for analysis.

Laboratory Analysis of Fish Samples

Fish samples were washed with distilled water and arranged on laboratory bench covered with aluminum foil. The wild species were identified using a fish guide (Paugy, Lévêque & Teugels , 2003). The body morphometrics such as Total Length and Standard Length was determined using a fish measuring board, while body weight was measured with a digital weigh balance (Adeogun *et al.*, 2020; Blankson, Tetteh, Oppong, & Id, 2022).

The measurements were followed by dissection of each fish (from the anal opening to the head region) for removal of the entire gastrointestinal tract (Blankson *et al.*, 2022). The gills were removed by opening the operculum; the gills were cut out with a pair of dissecting scissors. Guts and gills were weighed separately and cut into pieces into glass bottles. Digestion solution of (30 % 1:1 KOH:NaClO) was added to all the samples (5 mL of solution to 1 g of sample), based on the method of Enders, Lenz, Beer, & Stedmon, (2017). The sample was shaken in an electronic shaker to accelerate digestion. After the samples were digested, the digestate was emptied into a beaker, and floatation solution (hypersaline solution) was added (Adeogun *et al.*, 2020). The mixture was then stirred and allowed to settle for about 5 minutes, and the supernatant filtered through a membrane (pore size of 0.1 mm). The filtrate was rinsed with ethanol to facilitate drying at room temperature.

Dried residue from the membrane filter were picked unto a slide with forceps and examined under the Nikon SMZ-745 Stereo microscope and Euromex Nexius Zoom EVO Digital stereomicroscope with image analyses system camera for possible microplastics. The colour, size, shape and number of microplastics identified in each sample were recorded. These publications- Kuśmierek & Popiołek, 2020, Klangnurak & Chunniyom, 2020, Karbalaei et al., 2019 were used to classify the various MPs.

Sediment Sampling

Sediment samples were collected with a Van Veen grab from three different locations (upstream, midstream and downstream) at each of the sampling sites/communities. The sediment samples were separately wrapped in aluminum foils, and kept in labelled plastic ziploc bags and transported to the laboratory.

Laboratory Analysis of sediment

Sediment samples were oven-dried at 60 ˚C to constant weight. The dried sediment samples were homogenized using a laboratory ceramic pestle and mortar. Twenty (20 g) of the homogenized sediment was weighed into a glass beaker, and supersaturated NaCl solution (density ρ = 1.2 g/mL) was added. The supersaturated NaCl solution (ρ = 1.2 g/mL) is a separation fluid/solvent used to isolate microplastics from sediments where microplastic polymers of lower density float (Karlsson *et al.*, 2017). It is cost-effective and environmentally acceptable/friendly.

A drop of olive oil was added to the content (to improve recovery) using a dropper pipette and stirred using the magnetic stirrer for 10 minutes. The olive oil was added to prevent plastic particles from sticking to the glass walls, and subsequently collecting in the oil. The mixture in the glass beaker was covered with aluminum foil and then left for 4 hours to precipitate, after which the supernatant was slowly poured into the test tube. Organic material in the supernatant was digested by adding (30 %) H_2O_2 (3 mL H_2O_2 to 5 mL supernatant) into a glass tube, covered with a lid and left to react for about 20 minutes (Karlsson *et al.*, 2017). The digested supernatant was filtered through a filtrate material (pore size of 0.1 mm) and dried at room temperature. The wet hydrogen peroxide oxidation was applied to digest organic matter that can hamper the microplastic determination in the sample (Zobkov, Zobkova, Galakhina, & Efremova, 2020). (MPs extraction in sediment were done by density separation).

Dried residue from the filtrate material were picked unto a slide with forceps and examined under a Euromex Nexius Zoom EVO Digital stereomicroscope. The colour, size, shape and number of microplastics identified in each sample was recorded.

Water Sampling

Water samples were collected at sampling points (upstream, midstream and downstream) at each site using an 80 µm mesh conical net towed for 15-20 mins. The net was thoroughly rinsed with local/river water after each towing, and all collected materials were transferred to clean wide-mouth glass bottles, based on the modified method of Liu *et al.* (2021). The volume of water passing through the mouth of the net was estimated by the area of the conical net and the distance covered at each period of towing = net mouth area (m^2) × distance covered (m). Temperature, pH, conductivity and dissolved oxygen of the water at various communities were measured in situ with a Lovibond® Multiparameter kit.

Laboratory Analysis of Microplastic in Water

In the laboratory, 700 mL water sample was measured and the water filtered through a membrane/material with a pore size of 0.1 mm, with a vacuum pump (vacuum pressure < 2 Pa, Anti-Corrosion Diaphragm Vacuum Pumps-Hawach), based on the method of Liu *et al.* 2021. (MPs extraction in the water was done by density separation). The materials collected on the membrane were rinsed into a beaker using potassium hydroxide solution at a sample: solution ratio of 1:3 to digest any organic matter (10 % KOH was made with distilled water and filtered through a 10-13 µm (retention range) filter paper/membrane) (Liu *et al.*, 2021). The beaker was covered with aluminum foil and digested at 40 °C for ~ 48 h (Liu *et al.*, 2021). The digested liquids were then pumped through a membrane/material with a pore size of 0.1 mm. The membrane was then placed in a new petri dish and dried at room temperature. Dried residues from the membrane filter were picked unto a slide with forceps and examined under Euromex NexiusZoom EVO Digital stereomicroscope with image analyses system camera for possible microplastics. Colour, size, shape and number of microplastics identified in each sample was recorded.

Feed Sample Collection

Samples of feed used at the different farms were collected. Three brands of feed were obtained, coded as Feed A, Feed B and Feed C. Each feed collected was wrapped in aluminum foil and kept in labelled Ziploc® bags and brought to the laboratory for analysis.

Laboratory Analysis of Fish Feed

20 g of fish meal from each farm was weighed and transferred into a 500 mL DURAN glass bottle (Schott, Germany) sealed with a premium screw cap. Next, 200 mL (1:10 w/v) of KOH (10 % w/v) was added to each bottle, and incubated at 40 °C for 72 h (Karbalaei *et al.*, 2019). Digestates were filtered through 0.3 mm filter membranes using a vacuum pump (vacuum pressure < 2 Pa, Anti-Corrosion Diaphragm Vacuum Pumps-Hawach) connected to a filter

funnel manifold. To separate MPs from high density materials (shells and bones), the 0.3 mm filter membrane was immersed into 10-15 mL of NaI (4.4 M; 1.5 g/mL), sonicated at 50 Hz for 5 min and agitated on an orbital shaker at 200 rpm for 5 minutes (Karbalaei *et al.*, 2019). Finally, the mixture was centrifuged at 500xg for 2 minutes, and the supernatant containing MPs was vacuum filtered through a 10-13 μm (retention range) filter membrane (glass microfiber filter by CHMLAB Group, size = 47mm ⌀). This process was performed once more to ensure complete isolation of MPs.

Dried residue from the filtrate membrane were picked unto a slide with forceps and examined under the Nikon SMZ-745 Stereo microscope and Euromex Nexius Zoom EVO Digital stereomicroscope with image analyses system camera for possible microplastics. Colour, size, shape and number of microplastics identified in each sample was recorded.

Quality Assurance and Quality Criteria

Quality controls for the samples were conducted during the sampling and extraction process. Pure cotton clothes were worn during all sampling steps and laboratory procedures. All solutions, such as KOH:NaClO, KOH, H_2O_2, and distilled water, were filtered through a 10-13 μm (retention range) filter paper (150 mm in diameter). All lab supplies were rinsed three times with filtered distilled water before use, and dried on a clean test bench covered with aluminum foil. The laboratory work was conducted under a laminar-flow hood and use of plastic instruments or containers was excluded to the greatest degree possible throughout the experiments.

Data Processing and Analysis

The normality of variables was tested with Anderson Darling Test. For variables with a normal distribution, t student test and ANOVA were used, and when normality was not observed Welch's t -test and Kruskal-Wallis was used to compare between any 2 sets of groupings- cultured and wild source, and morphology types and abundance. Statistical significance was accepted at $p < 0.05$, and average values expressed as mean ± standard deviation on the mean (SD). Statistical analyses of the data were performed using Minitab Statistical Software and Excel.

Data was pooled for wild species at each site, independent of the season. The condition factor (K) and growth coefficient (b) were estimated from the length and weight relationship of the fish. The equation for the condition factor and growth coefficient is K=(100*W/Lb); where K indicates nutritional and physiological status of the fish. An estimated K greater than one (1) indicates good growth conditions, and $K < 1$ indicates poor growth conditions (Santos *et al.*, 2022). The value of the exponent b provides biological information on the kind/pattern of growth of fish. The growth is isometric if b=3, and the growth is allometric if $b \neq 3$ (negative allometric if $b < 3$ and positive allometric if $b>3$) (Santos *et al.*, 2022). Additionally, differences/variations in abundance and distribution of microplastic within the various samples for the different seasons, and sampling locations were analyzed using principal component analysis (PCA). The raw data was treated by cleaning, removing outliers and sorted before constructing the plots (Reid & Spencer, 2009).

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CHAPTER FOUR

RESULTS

General Morphometrics, Growth and Condition of Fish

A total of 330 fish samples were analysed in this study; 12 fish species were obtained from both wild and cultured sites. Of these, 23 represented single fish species (fish obtained from the wild for which comparisons could not have been made), 240 were cultured and 67 were from wild sites. For fish morphometric data, the standard length (SL) and weight were measured. Fish standard length ranged between 9.80 and 32.00 cm. Groupings of wild fish species showed standard length ranging between 9.80 and 21.30 cm for species- *Tilapia zillii*, *Synodontis vellifer*, and *Chrysichthys auratus*. A slightly higher standard length was obtained for *Tilapia guineensis*, *Brycinus brevis*, and *Parachanna obscura* (range: 14.00-22.10 cm). Other wild fish species had standard length ranging between 11.90 and 32.00 cm (*Mormyrus spp.*, *Chiloglanis occidentalis*, *Chrysichthys nigrodigitatus* and *Malapterurus electricus*). *Oreochromis niloticus*, the only cultured fish species, had standard length ranging between 14.50 and 26.50 cm. Fish species weighed between 40.00 and 850.00 g. The least weight was obtained from *Tilapia zillii* and *Synodontis vellifer* with a range from 40.00 to 220.00 g. Groupings of wild fish weight for species- *Parachanna obscura*, *Brycinus brevis*, *Chrysichthys nigrodigitatus*, and *Mormyrus spp*. ranged between 70.00 and 150.00 g. In order of weight, *Oreochromis niloticus* had the highest weight of - 850.00 g (range: 70.00-850.00 g), followed by *Malapterurus electricus*- 635.00 g (range: 410.00-635.00 g) and *Tilapia guineensis*- 460.00 g (range: 130.00-460.00 g). Fish species *Chrysichthys auratus* and *Auchenoglanis occidentalis* had weight ranging between 178.00 and 375.00 g. Fifty-eight (58 %) of the total species preferred a depth-integrated habitat in the water column, a few metres above the bottom (demersals); thus, feeding on MPs that settle at the bottom. The remainder of the species (42 %) were benthopelagics, that live within the water column, and feed on MPs that float within the column as well as those on the surface. About, 85 % of the total fish were benthopelagics. 50 % of the fish species were demersals from wild sites, 40 % were benthopelagics from the wild, and 10 % of the fish species were benthopelagics from cultured sites. Below, Table 1 shows the morphometric characteristics, growth coefficients, habitat and condition factors of the fish species for both cultured and wild fish used in the study. There were definite differences in weight and standard lengths between the cultured species obtained from the three sampling sites for the different seasons ($p < 0.05$). For the dry season, the mean weight and length of cultured species were higher for Kpong (μ_{weight} = 513.88 ± 114.32 g, μ_{length} = 21.96 ± 1.44 cm), in comparison to Sogakope Agordomi (μ_{weight} = 339.23 ± 64.76 g, μ_{length} = 19.36 ± 1.03 cm) and Asikuma Labolabo (μ_{weight} = 224.25 ± 53.85 g, μ_{length} = 16.67 ± 1.17 cm). In the wet season, the mean weight and length values obtained were (μ_{weight} = 447 ± 109.46 g, μ_{length} = 20.46 ± 1.6 cm), (μ_{weight} = 372.75 ± 32.75 g, μ_{length} =19.84 ± 0.49 cm), and (μ_{weight} = 366.25 ± 247.65 g, μ_{length}= 20.39 ± 3.50 cm) for Kpong, Sogakope Agodormi, and Asikuma Labolabo respectively. With wild fish species sampled in the dry season, species, such as *Tilapia guinessis*, *Mormyrus spp.*, *Chrysichthys nigrodigitatus* and *Synodontis vellifer* were obtained at Kpong and recorded mean length and weight as - (μ_{length} = 16.62 ± 2.77 cm, μ_{weight} = 236.15 ± 93.57 g), (μ_{length} = 23.70 ± 2.82 cm, μ_{weight} = 108 ± 21.39 g), (μ_{length} =17.96 ± 4.8 cm, μ_{weight} = 144.29 ±

132.76 g) and (μ_{length} = 15.6 ± 0.81 cm, μ_{weight} = 130 ± 127.28 g), respectively. *Chrysichthys auratus*, *Chrysichthys nigrodigitatus*, *Synodontis vellifer* and *Tiliapia zillii* were obtained from Asikuma Labolabo, and recorded respective mean length and weight as- (μ_{length} =14.76 ± 1.36 cm, μ_{weight} = 51.43 ± 11.80 g), (μ_{length} =13.93 ± 0.92 cm, μ_{weight} = 51.43 ± 11.80 g), (μ_{length} = 14.7 ± 4.38 cm, μ_{weight} = 51.5 ± 2.12.g) and (μ_{length} = 10.95 ± 1.6 cm, μ_{weight} =55 ± 21.21 g). For the wet season, *Chrysichthys nigrodigitatus* was the only wild species sampled at Sogakope Agodormi and recorded mean length and weight as 16.5 ± 0.79 cm and 96 ± 18.17 g. *Tilapia zillii* and *Parachanna obscura* were obtained at Kpong and had corresponding mean length and weight as (μ_{length} = 12.22 ± 0.73 cm, μ_{weight} = 99 ± 15.170 g) and (μ_{length} = 17.7 ± 1.00 cm, μ_{weight} = 77.5 ± 10.61 g). In Asikuma Labolabo, *Chenoglanis occidentalis*, *Brycinus brevis* and *Malapterurus electricus* were obtained and recorded mean weight and length as- (μ_{length} = 24.67 ± 2.31 cm, μ_{weight} = 310 ± 91.79 g), (μ_{length} = 18 ± 1.5 cm, μ_{weight} =110 ± 30 g) and (μ_{length} = 26.25 ± 3.18 cm, μ_{weight} = 522.5 ± 159.10 g), respectively.

The condition factor (K) describes the physiological and biological well-being of the fish. For *Oreochromis niloticus,* $K \geq 1$ indicated a good condition factor. Growth coefficient (b) described the length-weight relationship in the growth of the fish. Growth can be allometric (growth is not equal/proportional in all parts of the body). For the cultured fish (*Oreochromis niloticus*), b = 3 indicated isometric growth, b > 3 indicated positive allometric growth, b < 3 indicated negative allometric growth (Mortuza & Al-Misned, 2013). For both seasons, the condition factor and growth coefficient of the cultured species- *Oreochromis niloticus*- for all three sites indicated good condition ($k \geq 1$) and showed positive allometric growth, except for the wet season for which fish from Kpong showed a negative allometric growth (0.32) (

Table 1). The different wild species showed different condition factors and growth coefficients. Most of the species indicated good conditions with allometric growth (

Table 1).

Table 1: *Morphometric, Growth Coefficients and Condition Factors of both Cultured and Wild Samples for the Study.*

Species	Habitat	n	Source	Site	Season	Length/ cm	Weight/ g	b	K	Mean MPs/fish
Oreochromis niloticus	Benthopelagic	40	cultured	Sogakope Agodormi	dry	19.36	339.23	3.31	4.62	6.00
Oreochromis niloticus	Benthopelagic	40	cultured	Sogakope Agodormi	wet	19.84	372.75	2.77	4.77	7.05
Chrysichthys nigrodigitatus	Demersal	5	wild	Sogakope Agodormi	wet	16.50	96.00	3.34	2.12	5.20
Oreochromis niloticus	Benthopelagic	40	cultured	Kpong	dry	21.96	513.88	3.12	4.78	7.55
Tilapia guineensis	Benthopelagic	13	wild	Kpong	dry	16.62	236.15	2.58	4.99	4.92
Mormyrus sp	Demersal	5	wild	Kpong	dry	23.70	108.00	0.65	0.86	3.00
Chrysichthys nigrodigitatus	Demersal	14	wild	Kpong	dry	17.96	144.29	1.61	2.73	7.15
Synodontis vellifer	Benthopelagic	2	wild	Kpong	dry	15.60	130.00	2.23	3.20	1.00
Oreochromis niloticus	Benthopelagic	40	cultured	Kpong	wet	20.46	447.00	0.32	5.14	8.40
Tilapia zillii	Demersal	5	wild	Kpong	wet	12.22	99.00	2.19	5.42	9.00
Parachanna obscura	Demersal	2	wild	Kpong	wet	17.70	77.50	2.45	1.39	12.50
Oreochromis niloticus	Benthopelagic	40	cultured	Asikuma Labolabo	dry	16.67	224.25	3.13	4.76	7.00
Chrysichthys auratus	Demersal	4	wild	Asikuma Labolabo	dry	14.76	68.00	2.28	2.10	6.00
Chrysichthys nigrodigitatus	Demersal	7	wild	Asikuma Labolabo	dry	13.93	51.43	3.46	1.87	8.30
Synodontis vellifer	Benthopelagic	2	Wild	Asikuma Labolabo	dry	14.70	51.50	0.14	2.07	9.80
Tilapia zillii	Demersal	2	wild	Asikuma Labolabo	dry	10.95	55.00	2.65	4.10	8.00
Oreochromis niloticus	Benthopelagic	40	cultured	Asikuma Labolabo	wet	20.39	366.25	3.11	3.85	18.58
Chiloglanis occidentalis	Benthopelagic	3	wild	Asikuma Labolabo	wet	24.67	310.00	3.41	2.02	20.00
Brycinus brevis	Demersal	3	wild	Asikuma Labolabo	wet	18.00	110.00	3.35	1.85	14.00

Malapterurus electricus	Benthopelagic	2	wild	Asikuma Labolabo	wet	26.25	522.50	2.54	2.85	10.00

Habitat Source: https://tropicalfreshwaterfish.com/data/ecosystems/Volta.htm, field data (2022)

n= number of individual fish samples, b = growth coefficient, K = condition factor of fish, Weight/g = mean weight of each fish species in grams, Length/cm = mean length of each fish species in cm. Mean MPs/fish= mean number of MPs in each fish species, season = the season of sampling of the fish species, site= sampling site.

Abundance and Morphology of MPs in Fish

In this study, microplastics were detected in the gills and guts of fish species examined. Out of 240 cultured and 67 wild fish samples, 97.9 % (n = 235 samples) and 98.5 % (n = 66 samples) were respectively found to have accumulated microplastics. For the varied fish species, a total of 2,700 potential microplastic particles were identified under the optical microscope. In the dry season, a decreasing trend in MPs abundance was observed in cultured fish- Kpong recorded the highest number of 307 MPs (mean of 7.68 ± 4.17 items/fish), followed by Asikuma Labolabo with 276 MPs (mean of 6.9 ± 3.47 items/fish), and Sogakope Agodormi with a record of 238 MPs (mean of 5.95 ± 5.18 items/fish). A reverse trend was observed for the wild sites- Asikuma Labolabo recorded a higher abundance of 123 MPs (mean of 8.2 ± 3.08 items/fish) followed by Kpong- 170 MPs (average of 5.31 ± 4.25 items/fish).

Fish samples collected in the wet season from cultured sites showed a higher number of MPs consumed at Asikuma Labolabo (742 MPs, with an average of 18.48 ± 10.31 items/fish). About half of this amount was consumed by cultured fish in Kpong (342 MPS, average of 8.55 ± 6.99 items/fish). One third of this amount was consumed by cultured fish in Sogakope Agodormi (285 MPs with its average 7.13 ± 3.97 items/fish). The same trend was observed for MP consumption by wild fish followed by the same order as that for the cultured fish. In order of decreasing MP consumption, the observed trend is: Asikuma (121 MPs, average of 15.13 ± 7.41 items/fish) > Kpong (70 MPs with an average of 10 ± 4.83 items/fish) > Sogakope Agordomi (26 MPs and an average of 5.2 ± 3.19 items/fish). There were significant differences in MP abundance between the sites and source of fish for the two seasons ($p < 0.05$) (Fig 2 below).

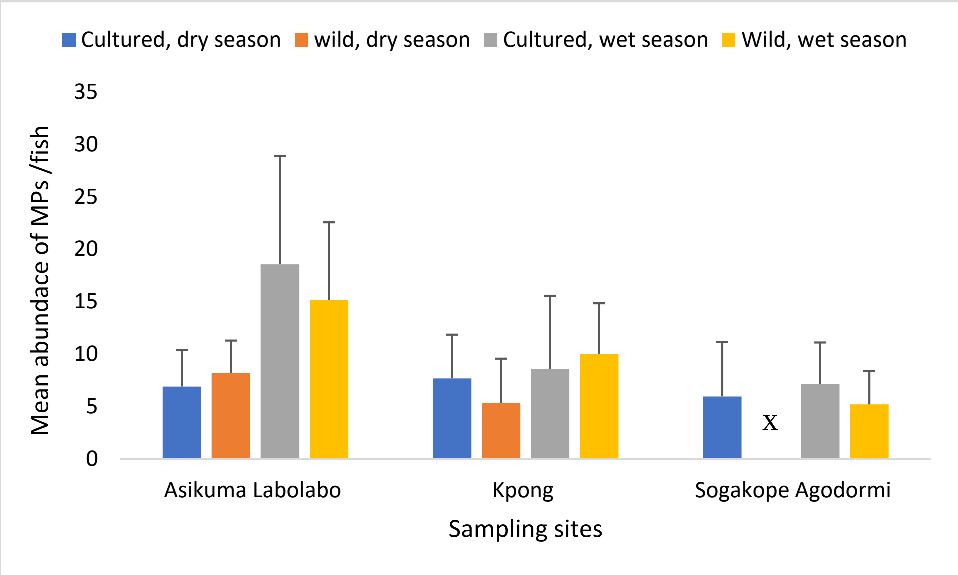

Figure 2: Mean abundance of MPs in fish sampled at cultured and wild sites (Asikuma Labolabo, Kpong, and Sogakope Agodormi) for the dry and wet seasons (cumulative) in the Lower Volta Basin.

x= samples were not taken from Sogakope Agodormi

The overall abundance of MPs in fishes for the 3 sites in the wet season (1586 MPs) was 1.4 fold higher than for the dry season (1114 MPs). In addition, the overall percentage abundance of MPs in both cultured and wild sites for both seasons was highest in Asikuma Labolabo (47 %), followed by Kpong (33 %), and Sogakope Agodormi (20 %); there were significant differences between the three sampling sites/locations (p < 0.05, Fig 3 below). At Kpong, the most microplastics extracted from an individual fish was obtained from the cultured (*Oreochromis niloticus*, µ=22 MPs) and wild site (*Chrysichthys nigrodigitatus*, µ=17 MPs) in the dry season. Some cultured species (*Oreochromis niloticus*) at Sogakope Agodormi and Asikuma Labolabo had no MPs. Also, no MPs were recorded in wild species (*Mormyrus spp.*) at Kpong. For the wet season, the highest abundance of MPs extracted from an individual cultured species, *Oreochromis niloticus* was 50 MPs; microplastics extracted from fish species from the wild site- *Chiloglanis occidentalis*- was 30 in total at Asikuma Labolabo. The least number for the cultured species was one (1 MP) at Sogakope Agodormi and Kpong, and one (1 MP) for wild species (*Chrysichthys nigrodigitatus)* at Sogakope Agodormi.

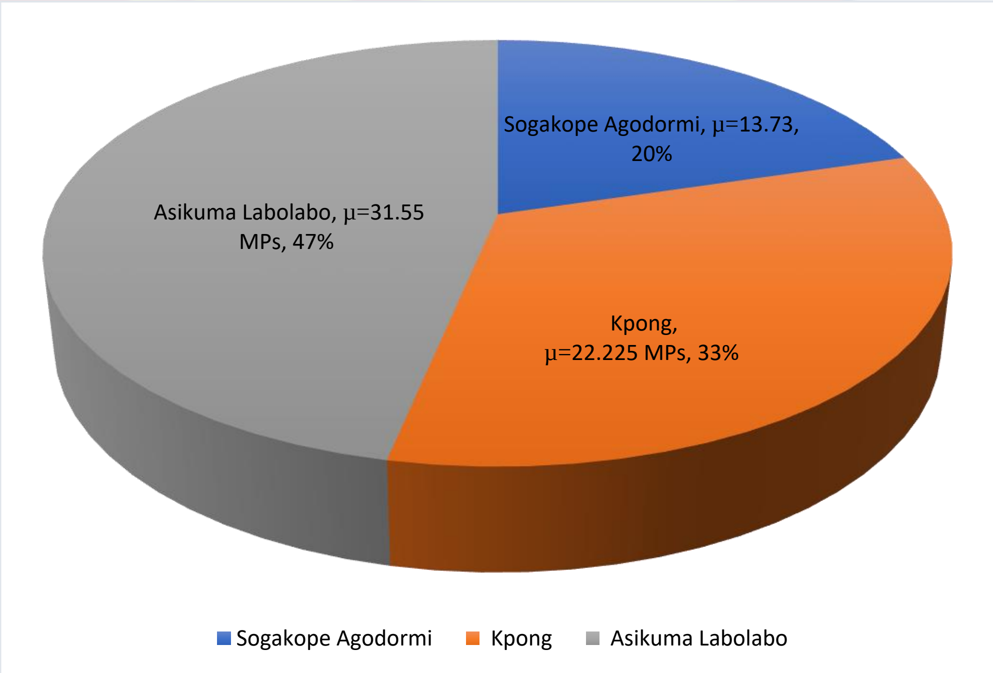

Figure 3: Mean(µ) and percentage abundance of MPs in fish (gut and gills-cumulative) at three sampling sites (Asikuma Labolabo, Kpong, and Sogakope Agodormi) for the dry and wet seasons (cumulative) in the Lower Volta Basin.

With reference to the two tissues analyzed, i.e. the gut and gill, the highest abundance of MPs identified in a single fish (cultured- *Oreochromis niloticus*) consisted of 27 MPs in its gut and 25 MPs in its gill at Asikuma Labolabo, while that for one fish sample taken from the wild site (*Chiloglanis occidentalis*) were 16 MPs (gut) and 20 MPs (gill) at the same location- Asikuma Labolabo. The least number for the cultured species were zero (0 MP) for the gut and one (1

MP) in fish gills at both Sogakope Agodormi and Kpong, respectively. No MPs were recorded in the gut and gill for wild species at Asikuma Labolabo. There were significant differences in MP abundance in the two types of tissues for each fish source for the three sampling locations ($p < 0.05$). In addition, the percentage (%) abundance of MPs per gram estimated was higher in the gills than guts for both cultured and wild species at all sites for both seasons. However, at Kpong the percentage abundances were equal in both the guts and gills for the wild species (Fig 4-6 below).

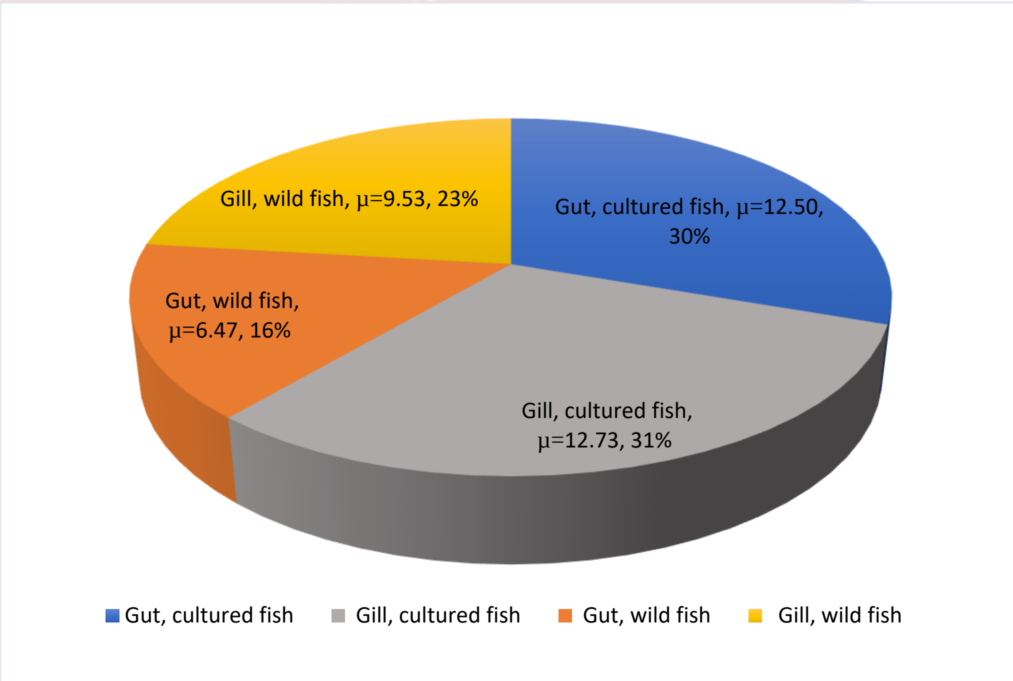

Figure 4: Percentage abundance of MPs in guts and gills of cultured and wild fish at Asikuma Labolabo for the wet and dry seasons (cumulative) in the Lower Volta Basin.

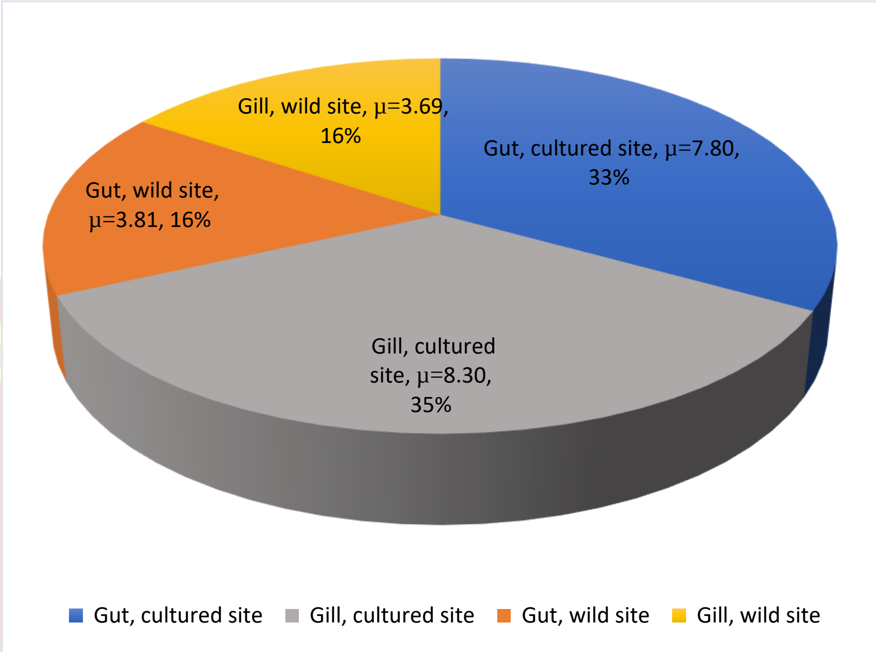

Figure 5: Percentage abundance of MPs in guts and gills of cultured and wild fish at Kpong for the wet and dry seasons (cumulative) in the Lower Volta Basin.

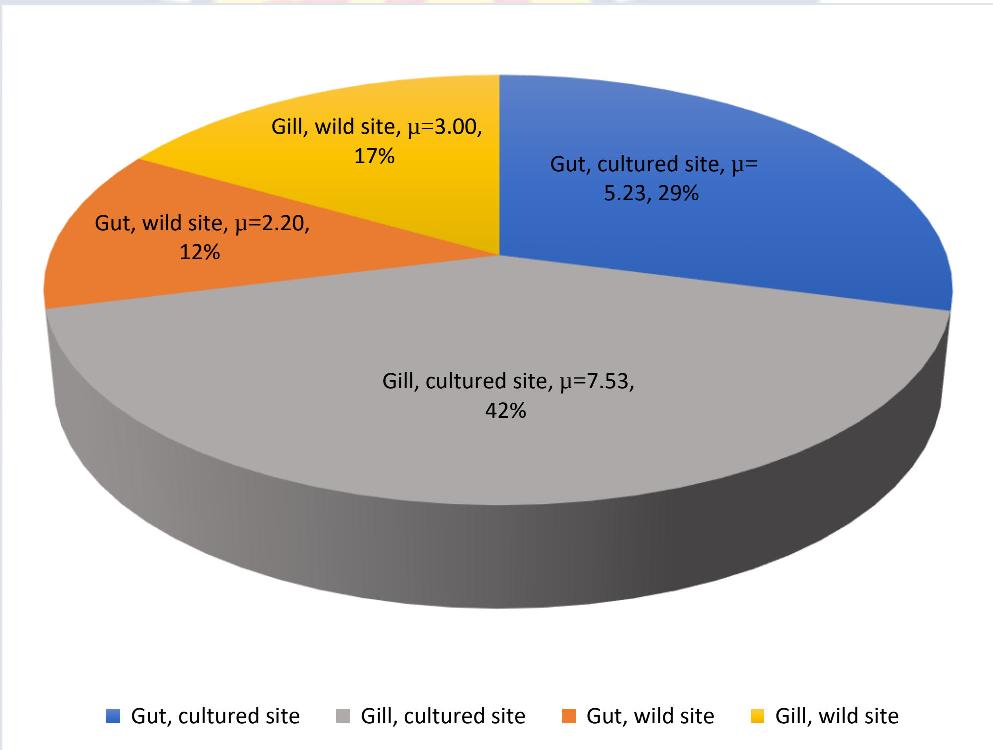

Figure 6: Percentage abundance of MPs in guts and gills of cultured and wild fish at Sogakope Agodormi for the wet and dry seasons (cumulative) in the Lower Volta Basin.

There are different shapes/types of MPs, and their distributions, were detected in fish source across the three sampling sites. Overall, the predominant MP shape identified were fibres (96.59 %); this was followed by fragments (3.30 %), and sheet/films (0.11 %) (Fig 7 below). Thus, fibre was the dominant MP type recorded for both dry and wet seasons for all three sampling sites. In the dry season, the abundant MP fibre (mean 11.5 ± 5.24 items/fish) was recorded at Kpong, while the least mean 5.73 ± 4.72 items/fish was recorded at Sogakope Agodormi. Kpong had high fragment abundance (mean 0.43 ± 0.93 items/fish), whiles Sogakope Agodormi recorded the least fragment abundance (mean 0.23 ± 0.70 items/fish). In the wet season however, Asikuma Labolabo recorded the highest fibre abundance (mean 20.8 ± 14.49 items/fish), whilst the least fibre (mean abundance of 7.5 ± 4.18 item/ fish) was recorded at Sogakope Agodormi. The highest fragment abundance was (mean 0.7 ± 2.23 items/fish) recorded at Asikuma Labolabo, while the least was (mean 0.28 ± 0.55 items/fish) recorded at Sogakope Agodormi. There were no sheet MPs recorded in the dry season at all three sampling sites. However, in the wet seasons, sheets were recorded only at Asikuma Labolabo with a mean abundance of 0.08 ± 0.35 items/fish. No beads or foam were detected in all 3 sites for both seasons. There were significant differences between the MP shapes at the sites for both seasons ($p < 0.05$) (Fig 8 below).

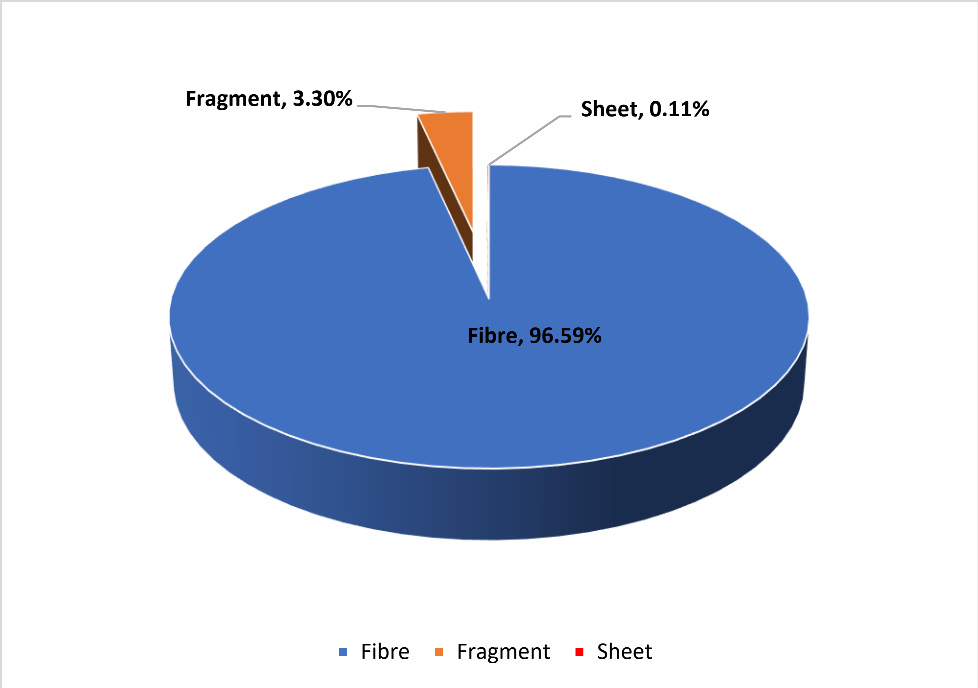

Figure 7: Percentage abundance of microplastic types (Fragments, sheets and fibres) extracted from fish sampled from the Lower Volta Basin.

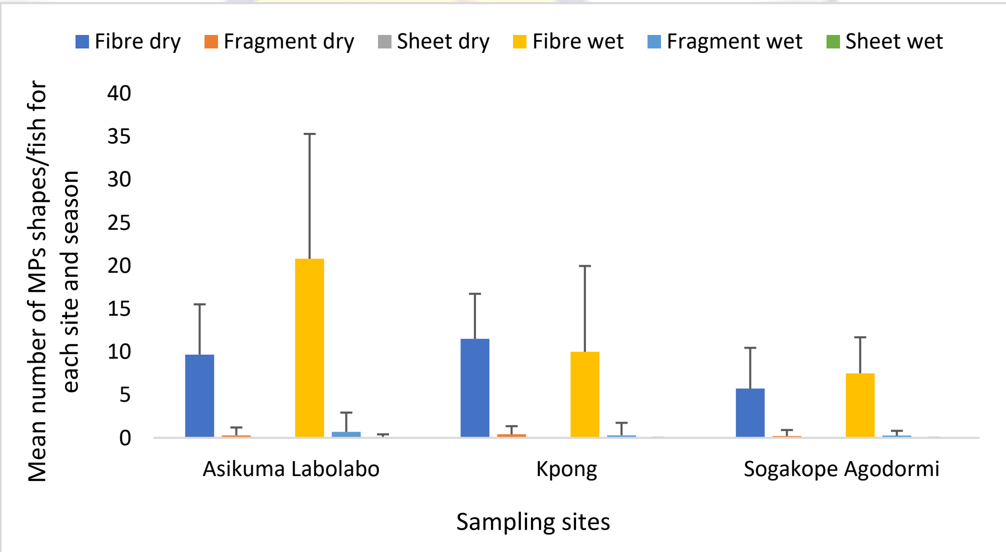

Figure 8: Microplastic types extracted from fish sampled from the Lower Volta Basin for the dry and wet seasons from wild and cultured sites within the 3 sampling sites (cumulative) (Asikuma Labolabo, Kpong, and Sogakope Agodormi).

Different colours of MPs were picked from fish for all sites, for both seasons. However, overall, black-coloured MPs (54 %) was the abundant, followed by blue-coloured MPs (19 %), red-coloured MPs (17 %) and cream-coloured MPs (5 %). These colours were identified in greater proportions than the rest of the other coloured MPs in fish from all 3-sampling sites (Fig 9 below). Other colours such as green, white and brown were identified but in very small percentages (all together 5 %) at some sites (Fig 10 below). For instance, brown-coloured MPs were identified in Kpong and Asikuma Labolabo, with a total of 11 MPs at both sites.

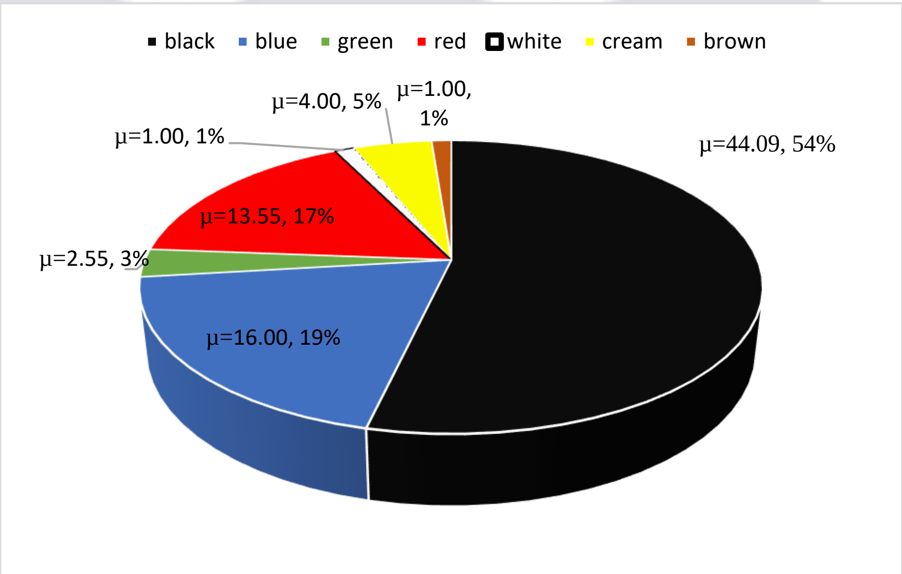

Figure 9: Abundance of microplastics extracted from fish across sites (Asikuma Labolabo-AL, Kpong-Kp, Sogakope Agodormi-SA) for the dry and wet seasons within the Lower Volta Basin.

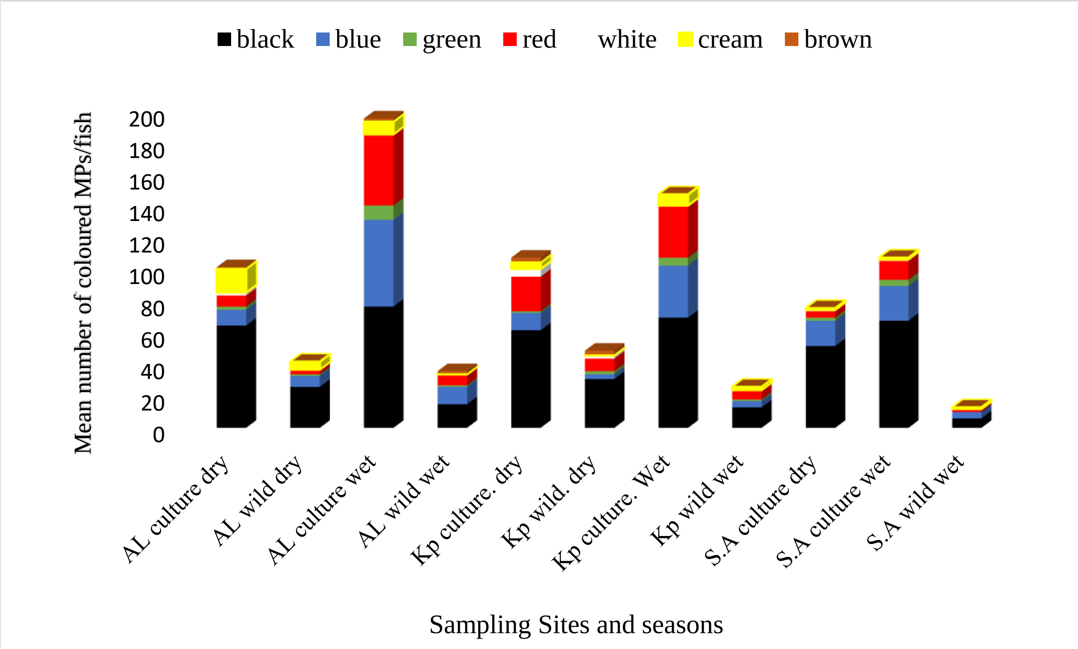

Figure 10: Mean and percentage abundance of coloured MPs ingested by fish from the 3 sites along the Lower Volta Basin for the dry and wet season.

The dimensions of the MPs were measured using a calibrated measuring slide underneath the microscope. Microplastics with size < 5 mm were of higher proportions in fish, at all sites for both seasons MPs in the size class of 0.51-1.00 mm were predominant (41 %), followed by 1.10-1.50 mm (20 %), < 0.50 mm (13 %), 1.51-2.00 mm (12 %). MP size class range of 2.10-2.50 mm, 2.51-3.00 mm (3 %), 3.10-3.50 mm (5 %), 3.51-4.0 mm (2 %), 4.10-4.5 mm (2 %), 4.51-5.0 mm (2 %) had smaller percentages, respectively (Fig 11below).

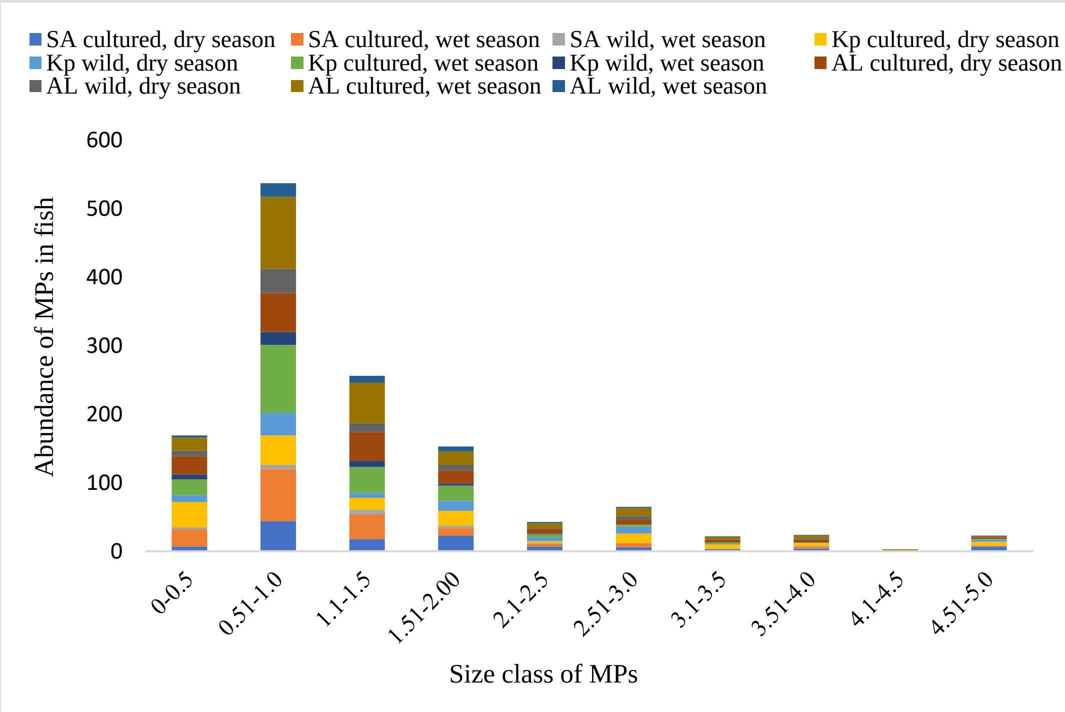

Figure 11: Microplastic size distribution based on their abundance in fish at different sampling sites (SA-Sogakope Agodormi, Kp-Kpong, AL-Asikuma Labolabo) and different sampling seasons.

Abundance and Morphology of Particles in Sediment

Microplastics were detected in all sediment samples from the three sampling sites, for both dry and wet seasons. In the dry season, the total MP abundance in sediment was high at Sogakope Agodormi - 459 MPs (mean: 153 ± 1.73 items/g), followed by Asikuma Labolabo- 381 MPs (mean :127 ± 90.02 items/g), and Kpong - 198 MPs (mean: 49.5 ± 44.59 items/g). Same trend was observed in the wet season, where Sogakope Agodormi recorded high MP sediment abundance of 487 MPs (mean: 162.33 ± 147.85 items/g), followed by Asikuma Labolabo- 288 MPs (mean: 96 ± 57.94 items/g), and Kpong- 137 MPs (mean: 34.25 ± 24.64 items/g). For both dry and wet seasons, the observed trend in order of decreasing mean abundance was Sogakope Agodormi > Asikuma

Labolabo > Kpong. There were no significant differences in MP abundance in the sediment at the three sampling sites, for the two seasons (T=1.16, p= 0.366) (Fig 12 below). With respect to the percentage abundance, for both seasons at each site, Sogakope Agodormi recorded high percentage MP abundance of 49 % followed by Asikuma Labolabo (34 %), and Kpong (17 %) (Fig 13 below).

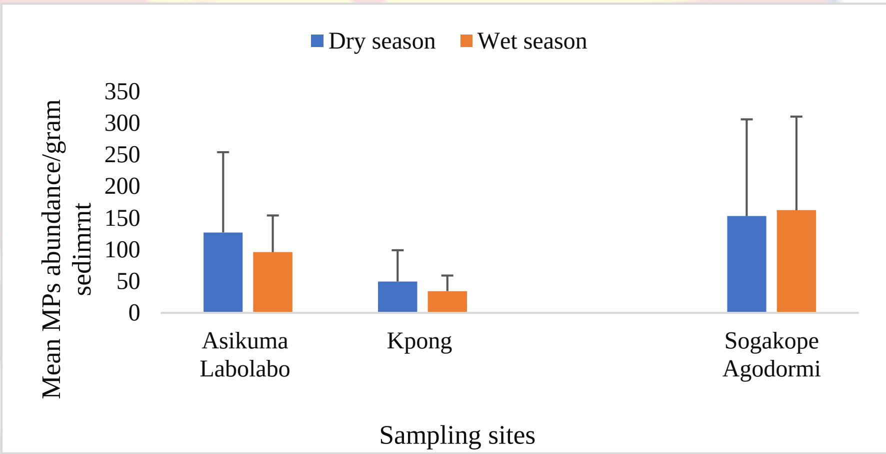

Figure 12: Mean MP abundance in sediment from the Lower Volta Basin at 3 sampling sites during the wet and dry seasons.

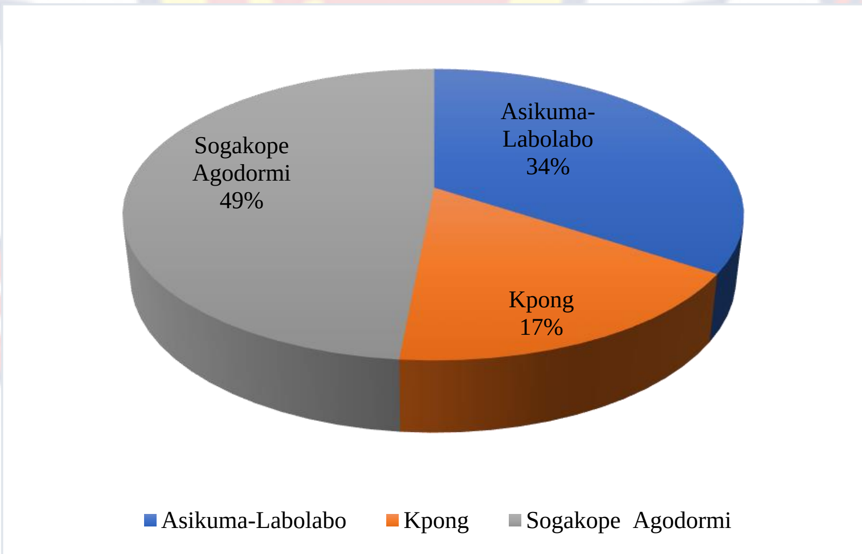

Figure 13: Percentage abundance of MPs in sediment from the Lower Volta Basin at 3 sampling sites- Sogakope Agodormi, Kpong and Asikuma Labolabo during the wet and dry seasons.

Also, the MP shapes in the sediment, fibres were abundant (99.08 %). This was followed by MP fragments (0.82 %), and sheets with a negligible percentage (0.10 %) (Fig 14 below). Furthermore, the highest mean fibre abundance in sediments was recorded at Sogakope Agodormi (dry season: μ=152.67 ± 1.53 items/g and wet season: μ=162.00 ± 147.37 items/g); while the least abundant (dry season: μ=48.00 ± 45.64 items/g and wet season:32.25 ± 25.75 items/g) were recorded at Kpong. The mean abundance of microplastic fragments were higher at Kpong (dry season: μ=1.5 ± 1.91 items/g and wet season: μ=2 ± 1.63 items/g), and the least was recorded at Asikuma Labolabo (No records of fragments for both seasons). In the dry season, MP sheets were not recorded at any site. However, Asikuma Labolabo was the only site in the wet season that recorded MP sheet with a mean abundance of 0.67 ± 1.15 (Fig 15 below).

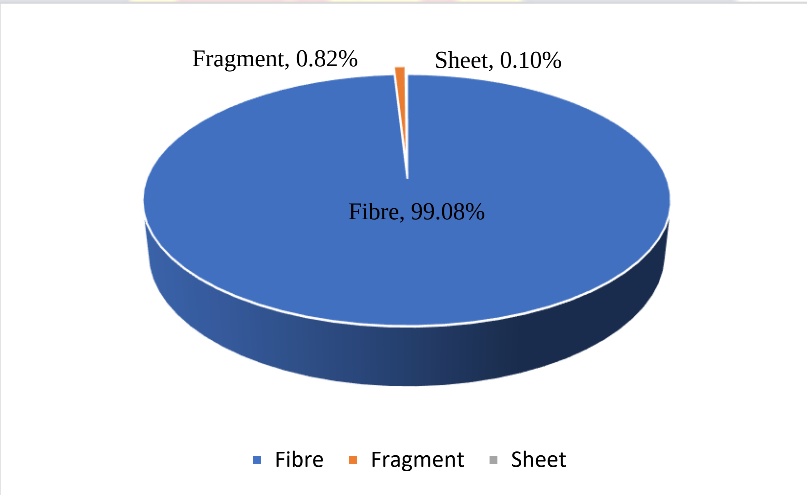

Figure 14: Percentage abundance of MP shapes in sediment from the Lower Volta Basin at 3 sampling sites- Sogakope Agodormi, Kpong and Asikuma Labolabo during the wet and dry seasons.

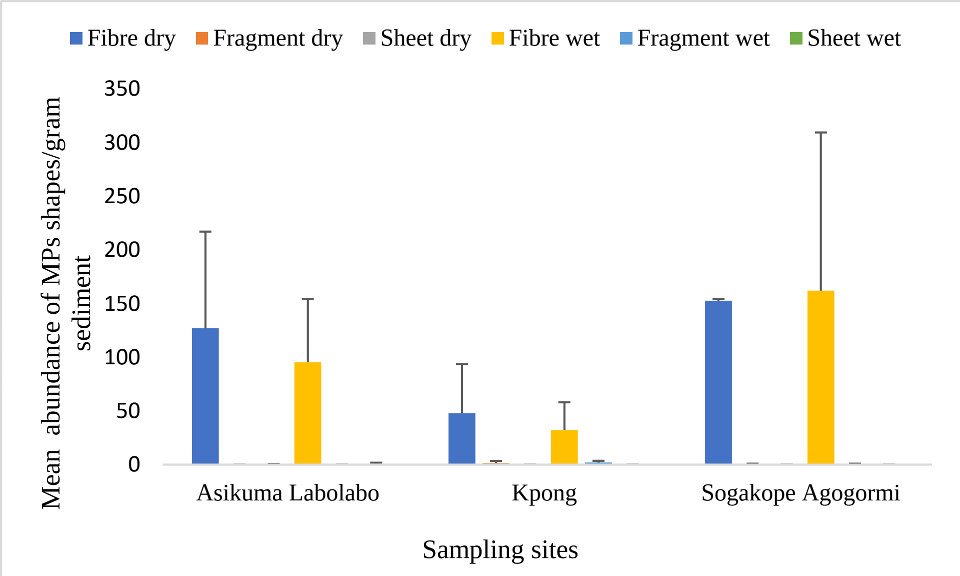

Figure 15: Mean abundance of MPs extracted from sediment from the Lower Volta Basin at 3 sampling sites in the wet and dry seasons.

Varied colours of MPs were obtained in sediment samples; this was same for MPs picked from fish, for all sites within both sampling seasons. Black-coloured MPs were the dominant (26 %); blue-coloured MPs contributed 22 %, 18 % of MPs were red in colour, and 13 % were contributed by white-coloured MPs (Fig 16 below). Other colours like green, cream, brown, yellow and transparent were identified but in small percentages (all together 21%). Black and blue-coloured MPs were the dominant microplastics that were consistently present at the 3 sites sampled, unlike for other coloured microplastics. The size of MPs ranged from 0 to 7 mm (longest length measured), with an average size of 2.05 mm. Length measurements of MPs in excess of 5 mm were eliminated, as these did not qualify as microplastics. About, 10 % of the microplastics were measured and dimensions reported. MPs in the size range from 0.51 to 1.00 mm were predominant (21 %). This was followed by those within the size range from 1.10 to 1.50 mm (16 %), 1.51-2.00 mm (14 %), and 3.51-4.00 mm (12 %). Microplastics of size < 0.50 mm, 2.10-

2.50 mm, 2.51-3.00 mm, 3.10-3.50 mm, 4.10-4.50 mm, 4.51-5.00 mm were in small counts, with percentages of 0 %, 8 %, 9 %, 8 %, 6 % and 6 %, respectively (Fig 17 below).

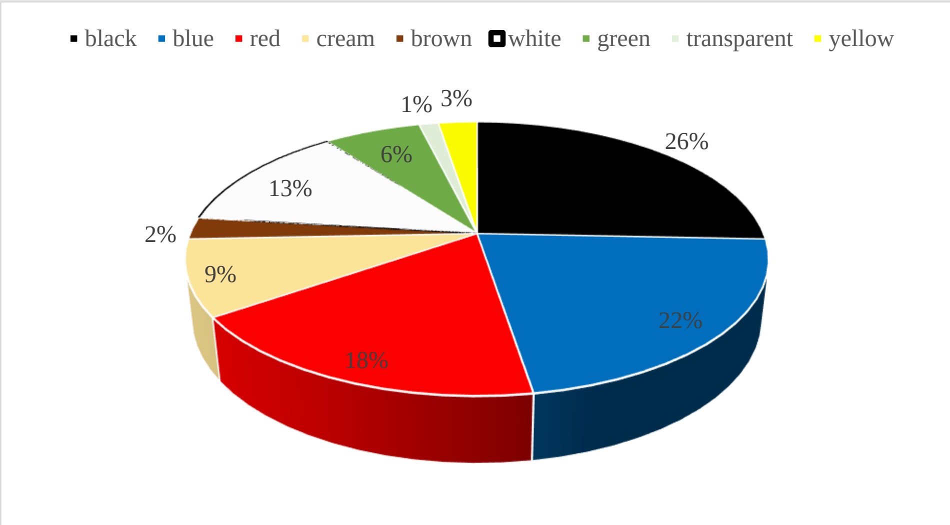

Figure 16: Percentage abundance of coloured MPs in sediment for 3 sampling sites along the Lower Volta Basin.

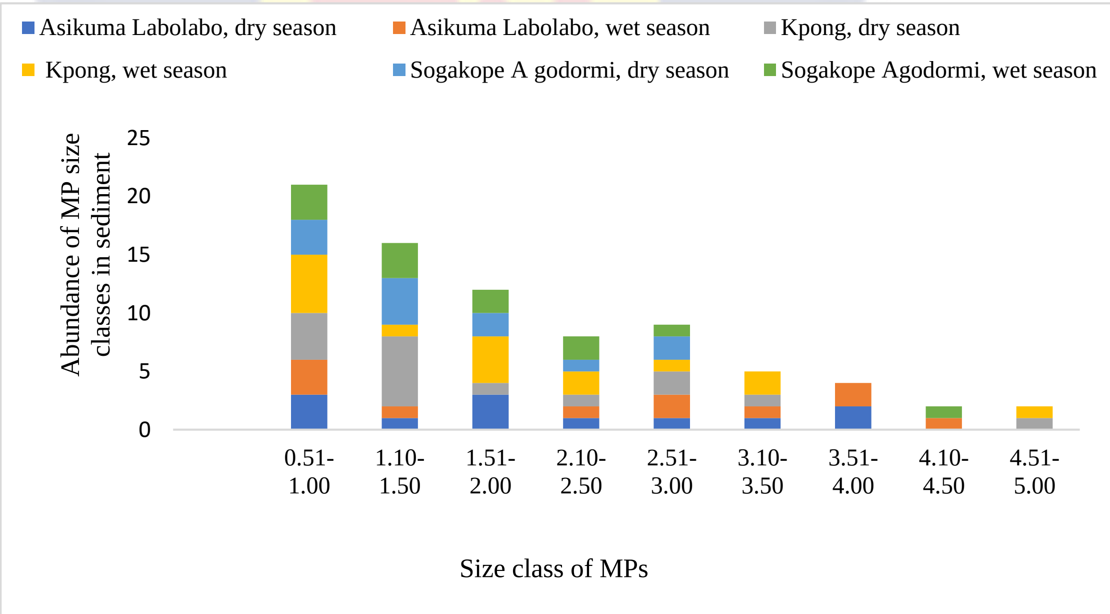

Figure 17: Microplastic abundance based on the size range in sediments from the Lower Volta Basin sampled from 3 sites within the dry and wet seasons.

Abundance and Morphology of Particles in Water

MPs were identified in all water samples from the three sampling sites. A total of 111 MPs were picked (mean: 37 ± 14.93 items/L). With respect to MP abundance recorded at the individual sampling sites, there was a decreasing trend from Sogakope Agodormi (48 MPs, mean- 16 ± 19.05 items/L) to Asikuma Labolabo (43 MPs, mean- 14.33 ± 8.50 items/L), and Kpong (20 MPs, mean- 5 ± 0.82 items/L). Thus, Sogakope Agodormi recorded the highest, and Kpong the least. This trend is similar to that observed in MP abundance in sediment in the wet season. There were no significant differences in MP abundance in water at the three sampling sites (T= 0, p = 1.0) (Fig 18 below).

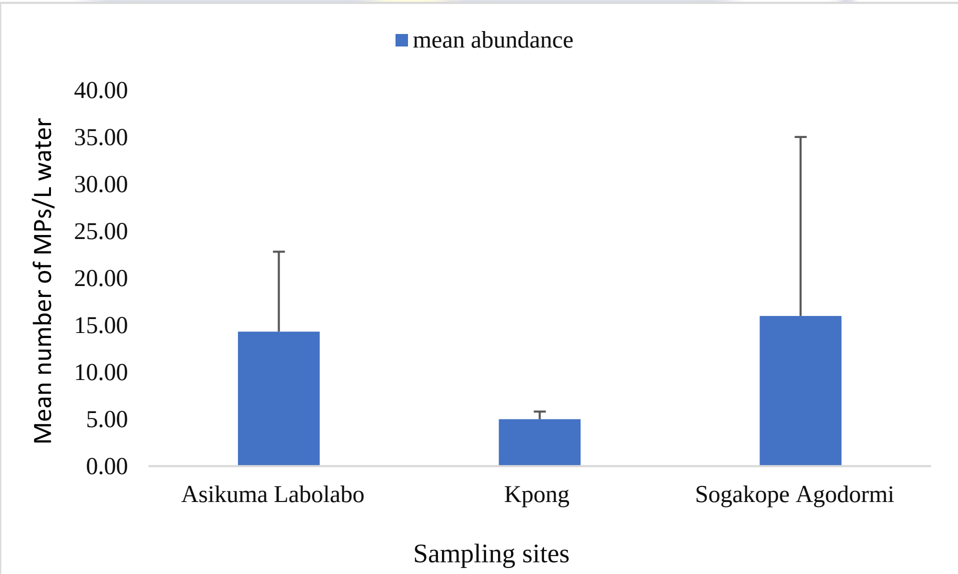

Figure 18: Mean MP abundance in water from the Lower Volta Basin sampled at 3 sites in the wet seasons.

Furthermore, the MP shapes observed in fish, sediment, and fibres were equally the most abundant MPs in water. The highest MP abundance was recorded at Sogakope Agodormi, with a mean of 15 ± 17.32 items/L; the least MP was reported at Kpong with a mean of 4.5 ± 1.29 items/L. The highest abundance of fragment-shaped MPs was recorded at Sogakope Agodormi (mean 1 ± 1.73), and the least abundant (mean 0.5 ± 1.00 item/L) at Kpong. There were no significant differences in MP shapes between the 3 sites (T=3.84, P=0.062) (Fig. 19 below). No sheets, bead or foam were recorded in the water samples.

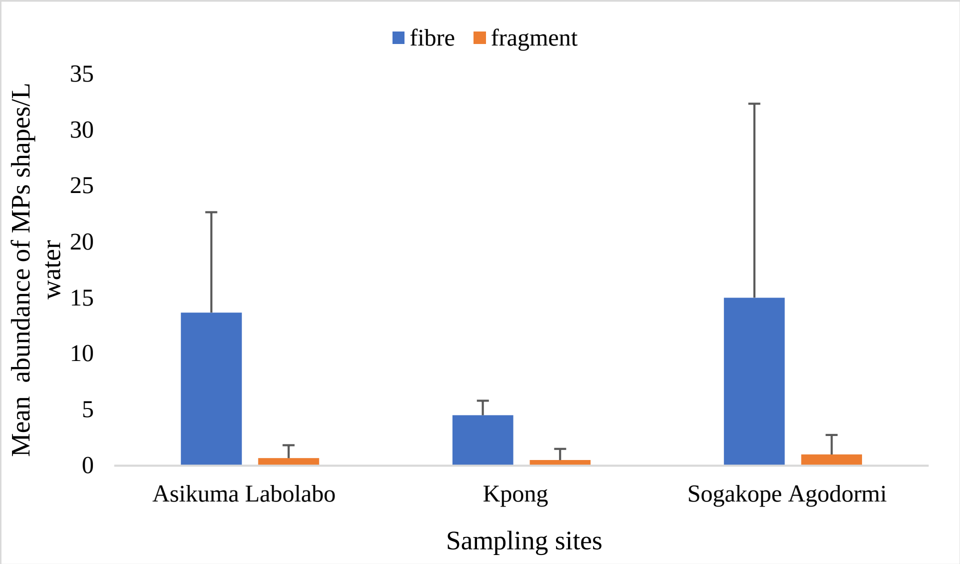

Figure 19: Mean abundance of MP shapes in water from the Lower Volta Basin collected from 3 sites in the wet season.

Black, blue, red and cream, were the only coloured MPs picked from the water samples. Blue coloured MPs were the most abundant (41 %). This was followed by red-coloured MPs (23 %); cream and black-coloured MPs, contributing a total of 18 %. (Fig. 20 below). Apart from black, all other coloured MPs were observed at all sites.

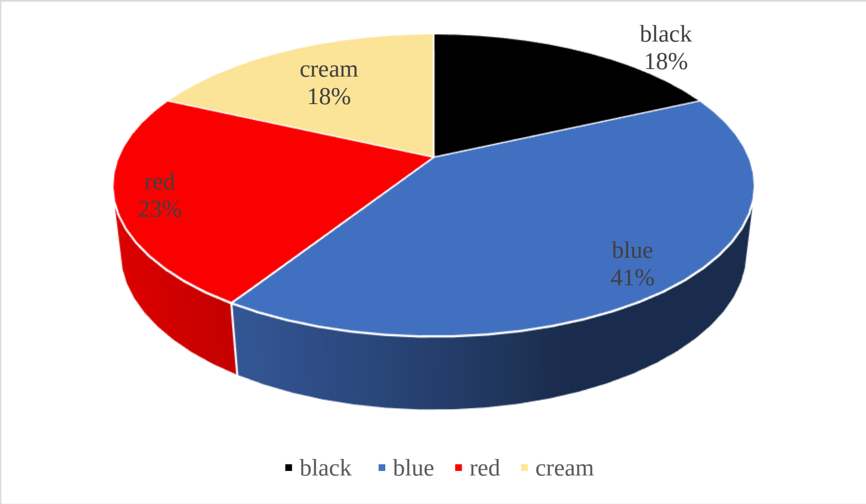

Figure 20: Percentage abundance of MPs colours in water from the Lower Volta Basin sampled at 3 sites in the wet season.

Similar to fish and sediment samples, the maximum length of MPs measured in water was 5 mm. However, a large portion of MPs < 5 mm were measured in all water samples. Microplastic size class range between 3.51-4.00 mm were predominant (31 %). This was followed by MPs with size ranging between 4.51-5.00 mm (21 %), and 1.51-2.00 mm (17 %). A lesser cumulative contribution of 7 % was obtained for microplastic size class ranging between 0.51 and 1.00 mm, 2.51 and 3.00 mm, and 4.10 and 4.50 mm. Much lesser contributions of 3 % was obtained for microplastic size ranges between 2.10 and 2.50 mm, and 3.10 and 3.50 mm. Microplastic sizes ranging between 1.10 and 1.50 mm contributed 4 % to the total MPs identified in water (Fig 21 below). Larger sized microplastics were more dominant in fish (67.57 %) in comparison to those detected in water (22.97 %) and sediment (9.46 %) for sizes ranging between 3.51 and 5 mm.

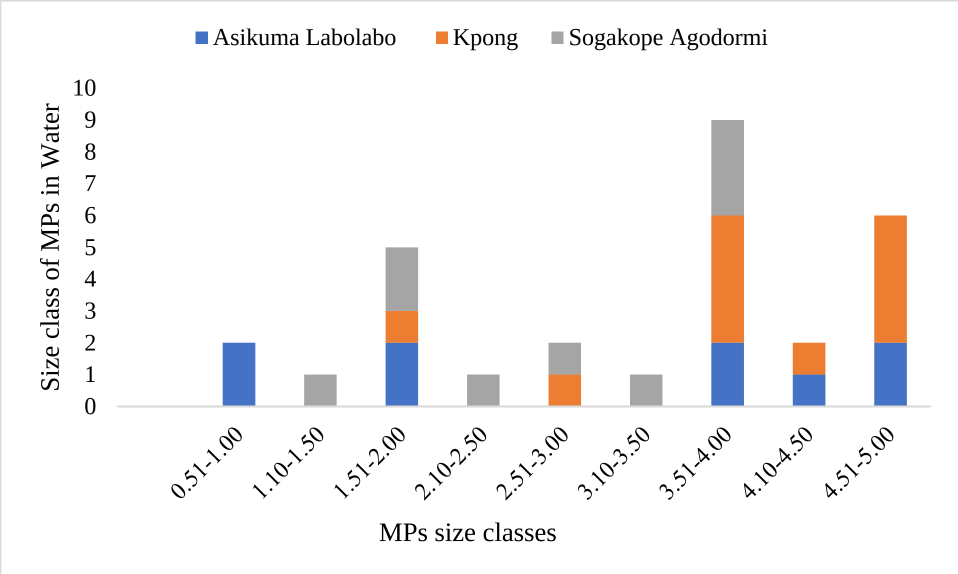

Figure 21: Abundance of MPs with respect to their size distribution in water from the Lower Volta Basin at 3 sampling sites for the wet season.

Abundance and Morphology of Particles in Feed

Microplastics were counted in all three fish feed analyzed, with the highest number counted in Feed A (75 MPs), and a mean of 25 ± 40.71items /g. This was followed by Feed B (47 MPs), with a mean of 15.67 ± 18.45 items/g. Feed C had the least number of microplastics: 20 MPs (mean 6.67 ± 11.55 items/g); thus, the order of abundance was Feed A > Feed B > Feed C (Fig 22 below). The difference in abundance was not significant among the three feed (T = 0, p =1.00).

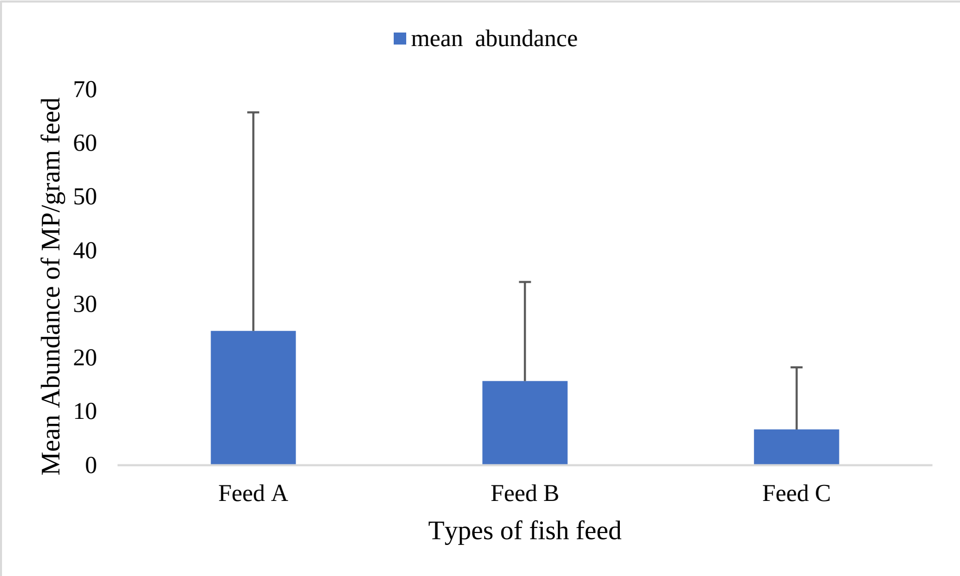

Figure 22: Mean abundance of microplastics identified in three fish feed.

Microplastic shape observed were fibres, fragments and sheets; there were no beads or foam MPs. Fibres were the dominant MPs detected. The total fibre from all three feed was 128 MPs with a mean value of 42.67 ± 26.63 items/g, fragments contributed 13 MPs (mean: 4.33 ± 5.86 items/g), and sheet 1MP (mean: 0.33 ± 0.58 items/g) (Fig. 23 below). Significant differences were observed among the MP types in the feed ($p < 0.05$).

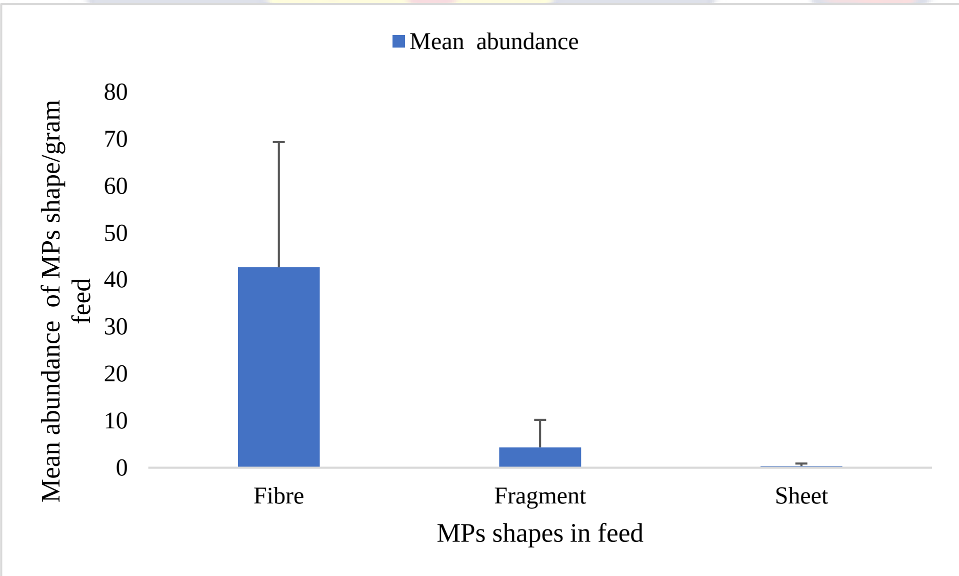

Figure 23: Mean abundance of microplastic shapes identified in three fish feed.

The colours of MPs observed in the feed were black, red, violet, yellow, orange, blue and white. Black was the most abundant (28 %) followed by red and blue (18 % each), yellow, orange, violet and white, likewise, were of equal abundance of 9 % each (Fig. 24 below).

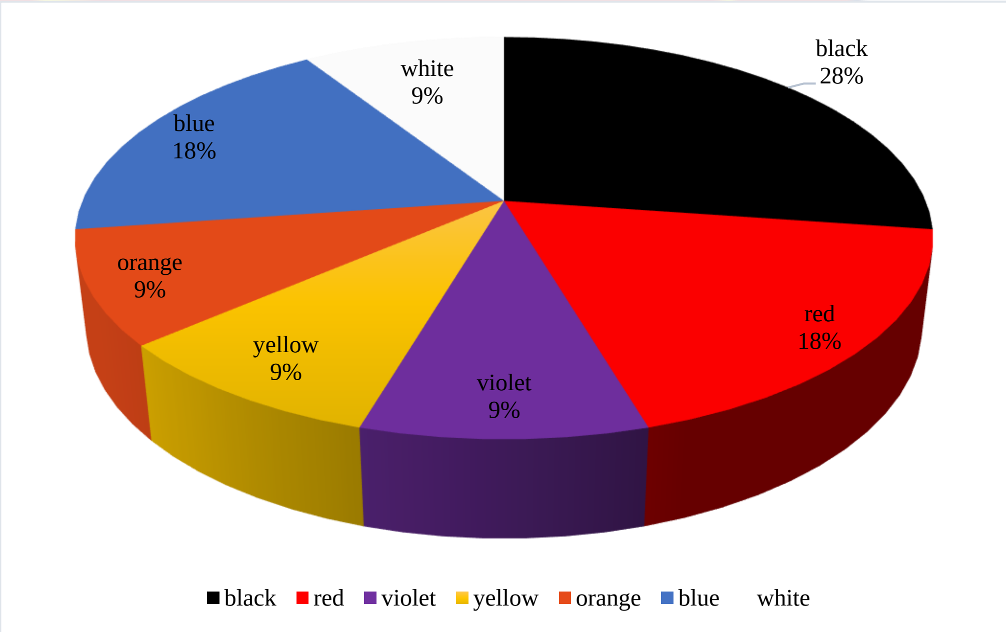

Figure 24: Percentage abundance of coloured MPs colours in fish feed.

The maximum length of MP measured in the feed was < 5mm. Unlike other sample types, just a few size classes of MPs were recorded in the feed. These ranged between 0.51 and 1.00 mm (41 %), 1.51 and 2.00 mm (25 %), 2.10 and 2.50 mm (17 %), and 3.51 and 4.00 mm (17 %) (Fig. 25 below).

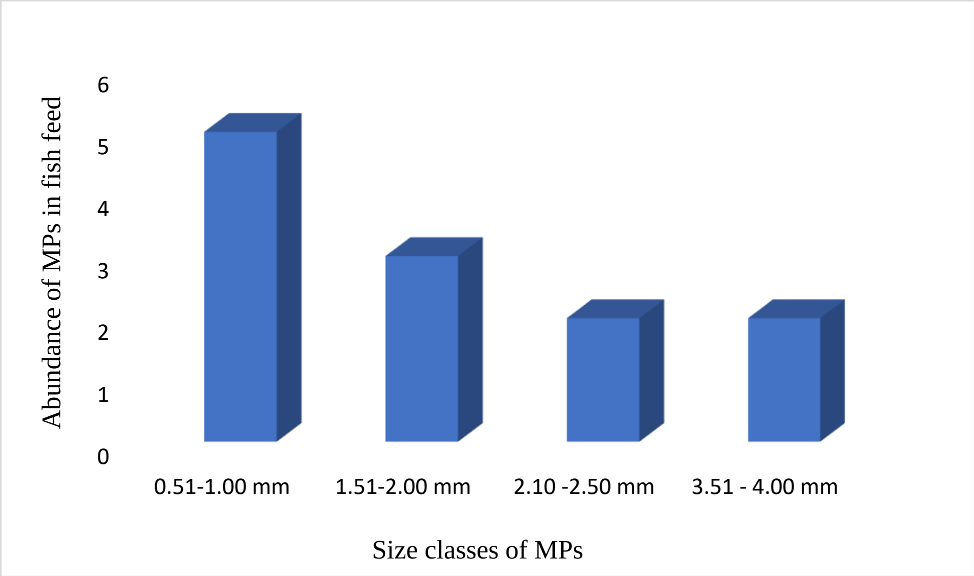

Figure 25: Microplastic abundance and their size classes in fish feed.

Total MPs Abundance at Each Site

The overall mean MP abundance in each of the sample type i.e. fish, sediment and water, at each sampling site, was in the order of Asikuma Labolabo > Sogakope Agodormi > Kpong (Fig 26 below). Thus, Asikuma Labolabo recorded the highest count of MPs; Kpong recorded the least. There was no statistically significant difference in MPs abundance between the three sample sites ($p > 0.05$).

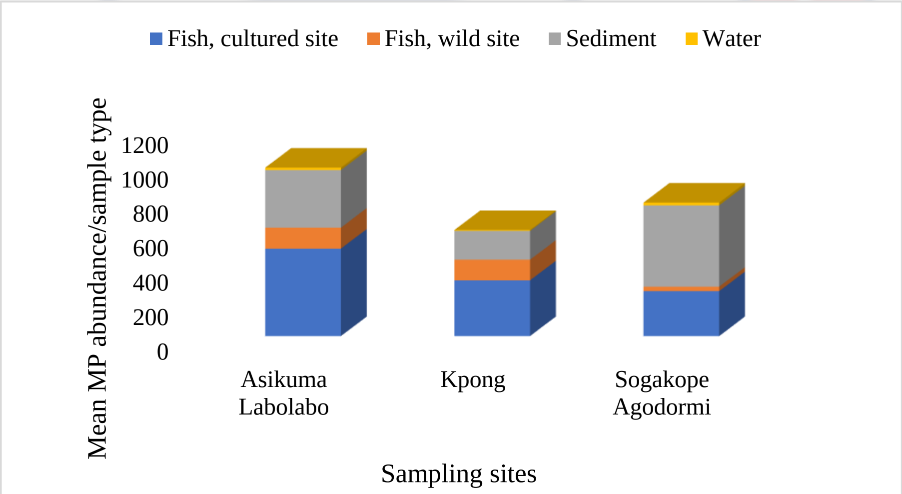

Figure 26: Mean abundance of MPs at each sampling site.

Principal Component Analysis for Fish Samples

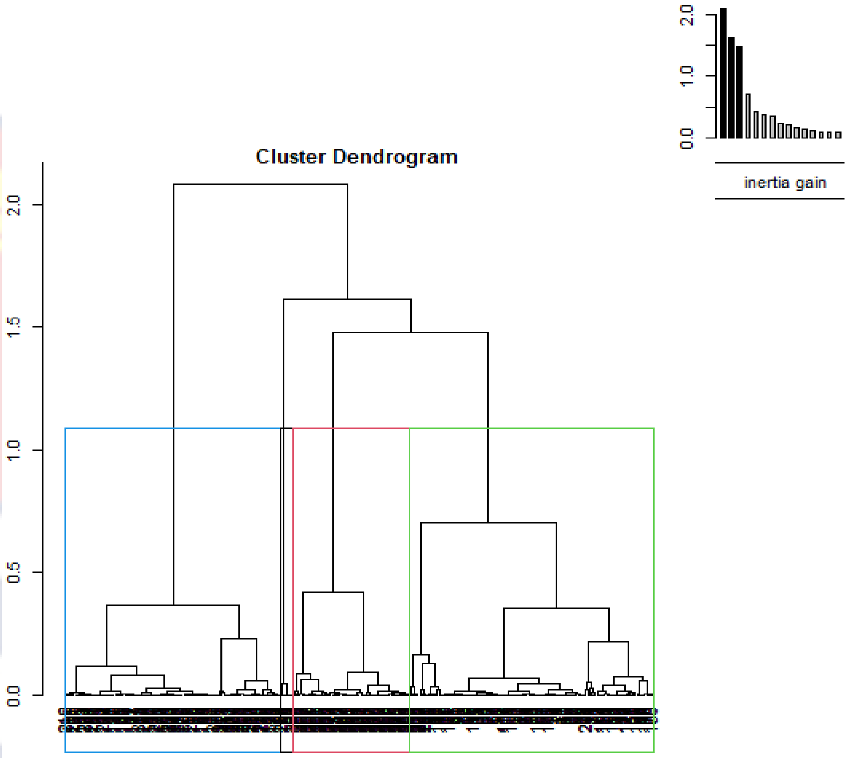

Figure 27: Hierarchical tree of clusters obtained from variables of MPs data in fish.

From the cluster analyses, concentrations of microplastics, their colour, size, shape, and sampling seasons, sampling sites, farmed and wild fish types, were grouped into 4 statistically significant clusters via a dendrogram. These plots are shown in Fig 27 above. Cluster 1 consists of samples from Asikuma Labolabo and Kpong. This clustering is characterized by a high MP abundance in fish sampled in the dry season; their size ranges from 0.51 to 1.50 mm. These consists of microplastic fibres of typical colours black, and blue. A minor contribution of fish sampled from the wet season contributed red-coloured microplastics of size ranging between 0- and 0.50 mm. Cluster 2 is made up of individuals that have high values for variables sampled during the dry season from wild fish. Distinctively, these microplastics have a size range between 1.51 and 2.00 mm; samples are drawn from all 3 sites: Asikuma, Kpong and Sogakope. Samples collected during the wet season from cultured sites did not contribute much to this cluster. Cluster 3 is characterized by contributions consisting of variables collected during the dry season from cultured fishes. Microplastics of size range between 2.51 and 3.00 mm, and 4.51 and 5.00 mm of brown colour in the form of sheets and fibres make up this cluster. Varied variables contribute to cluster 4. For cultured fish samples collected in the wet season, fibrous black, blue and red-coloured microplastics of size range between 0.51 and 1.50 mm, and 4.10 and 4.50 mm make up this cluster.

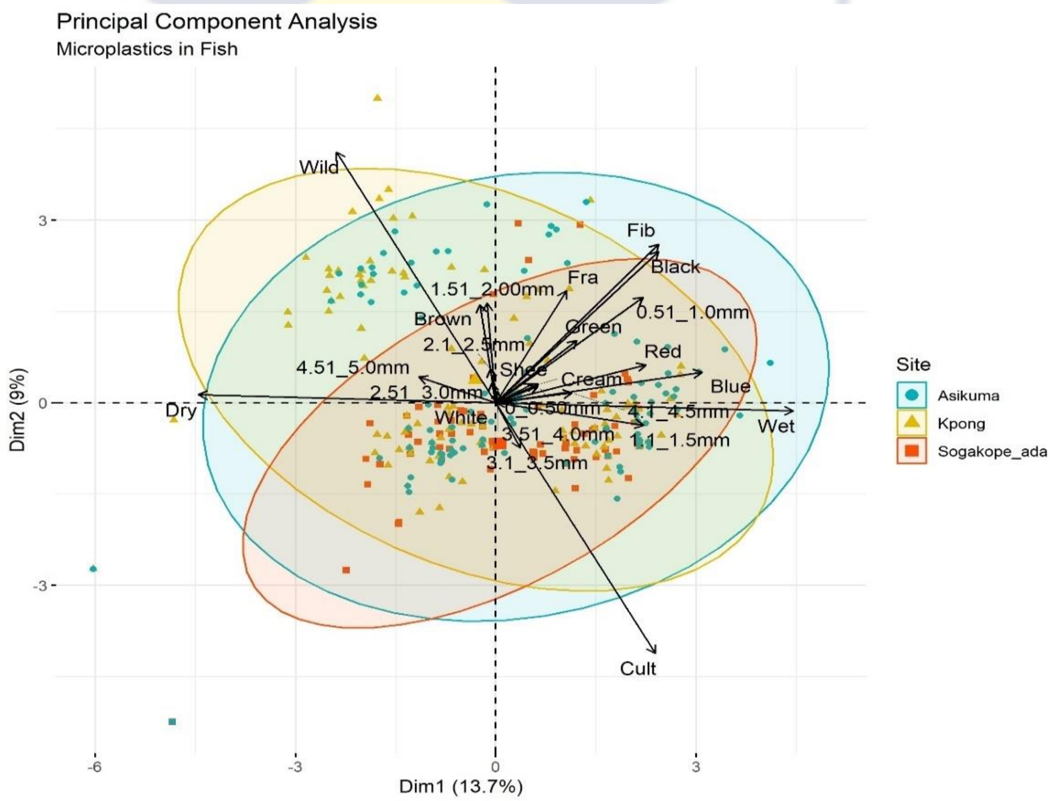

Figure 28: MPs variables in cultured and wild fish species from Asikuna Labolabo, Kpong and Sogakope Agodormi. (Dim: dimension/components).

Exploratory data analysis involving Principal Component Analysis (PCA) was used to investigate how the relative proportions of MP abundances in fish, water and sediment changed in different sampling sites. This was performed to determine if variables such as the season, sample matrices (fish species, water and sediment), MP size, shape and colour had distinct variations. A plot was constructed using R Version 4.2.3 to explain the observed relationships between sampling sites and MP variables. A consideration of all sample variables showed that two dimensions described the MP distributions. Dimension 1 was influenced by MPs ingested by cultured species (*Oreochromis niloticus*-benthopelagics) in the wet season. Most of the cultured species were from Asikuma Labolabo and Sogakope Agodormi. In *Oreochromis niloticus*, MPs ingested were blue, red and black fibres in size ranging between 0.51 and 1.00. mm.

A secondary group of MPs contributing to this dimension were those from cultured species -*Oreochromis niloticus* sampled in the dry season, from Sogakope Agodormi. Microplastics from these fishes were black-coloured, fibrous in nature, and small-sized- measuring between 0.50 and 1.00 mm. Other minor MP contributions sampled from wild fish- *Mormyrus sp*- in the wet season were blue and red coloured, with size ranging between 0 and 0.50 mm, and 1.51-2.00 mm. A third group of this dimension were MPs ingested by wild species in the dry season. These were mostly sampled from Kpong and Asikuma Labolabo. MPs observed were black and brown fibres of dimension ranging between 1.51 and 2.50 mm, and between 4.51 and 5.00-mm. Minor contributions to this group were blue MPs from cultured species sampled in the wet season; these ranged between 1.10 and 1.50 mm.

Dimension 2 was influenced by MPs ingested by wild species including *Chrysichthys nigrodigitatus* (benthopelagic), *Chiloglanis occidentalis* (benthopelagic), *Tilapia Zillii* (demersal), *Synodontis vellifer* (benthopelagic) etc. sampled from Kpong and Asikuma Labolabo. Black and brown coloured fibrous MPs were ingested by these wild species.

A second group of this dimension consisted of MPs from cultured species in the dry season from Sogakope Agodormi. Microplastics from these fishes were small-sized, black-coloured and measured between 0.50- 1.00 mm. In addition to MPs in this group were blue and red coloured ones measuring between 0-0.50 mm, and 1.51-2.00 mm ingested by the wild species- *Mormyrus spp.* in the wet season. A third group of this dimension consisted of MPs ingested by cultured species (*Oreochromis niloticus*-benthopelagic) in the wet season. Most of the cultured species here were from Asikuma Labolabo and Sogakope Agodormi. MPs ingested were black fibres in small size class of 0.51 to 1.00. mm. Fibrous MPs were more in fishes from these sites because of the high activities of cage farming in these areas. *Tilapia zillii*- a demersal wild species sampled from Kpong contributed blue and red coloured MPs. Sizes of MPs ingested by wild species at this site were both small-sized of dimensions ranging between 1.51-2.00 mm and big sized- 4.51-5.00 mm. In this dimension, most of the cultured species ingested small sized MPs, while wild species ingested different size range of MPs. Thus, MPs in fish were influenced by the different size classes of the MPs.

Principal Component Analysis for Water Samples

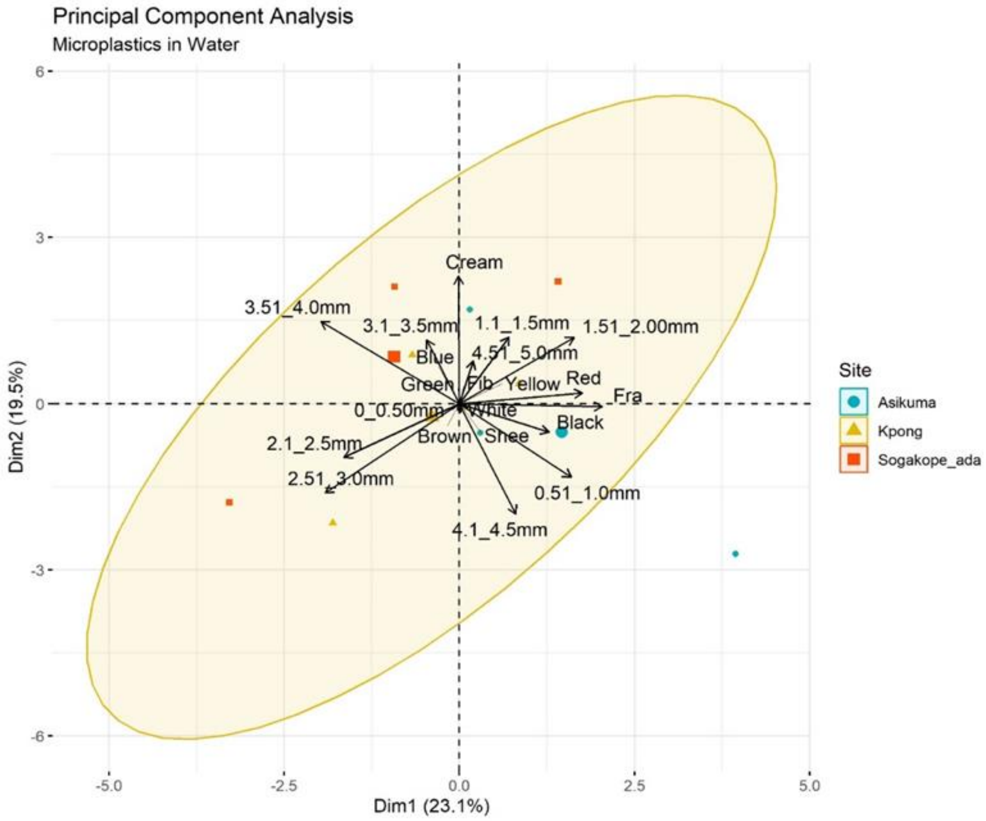

Figure 29: MPs variables in water from Asikuma Labolabo, Kpong and Sogakope Agodormi. (Dim: dimension/components).

With respect to water (Fig 29, above) Dimension 1 was influenced by small (0.51-1.00 mm) and large size MPs (4.1-4.5 mm). Few MPs in the medium size of 3.51-4.00 mm contributed to the group. MPs in this group were sampled from all three sites. A second group of MPs in the medium size class of 2.51-3.00 mm and 2.10-2.50 mm contributed to this dimension and were samples from Sogakope Agodormi and Kpong.

Dimension 2 was influenced by medium size class MPs of 2.51-3.00 mm to large size class of 4.1- 4.50 mm and 4.51-5.00 mm sampled from all three sampling sites. A second group of this dimension were MPs in medium size class of 2.51-3.00 mm and 2.10-2.50 mm from Sogakope Agodormi and Kpong.

Contributing to this dimension was a third group of MPs of small size class of 0.51-1.00 mm and large size class of 4.10-4.50 mm from Asikuma Labolabo and Kpong. Few medium size classes of 3.51-4.00 mm were part of the group. From both dimensions, MPs in water of all three sites were mostly medium to large size classes.

Principal Component Analysis for Sediment Samples

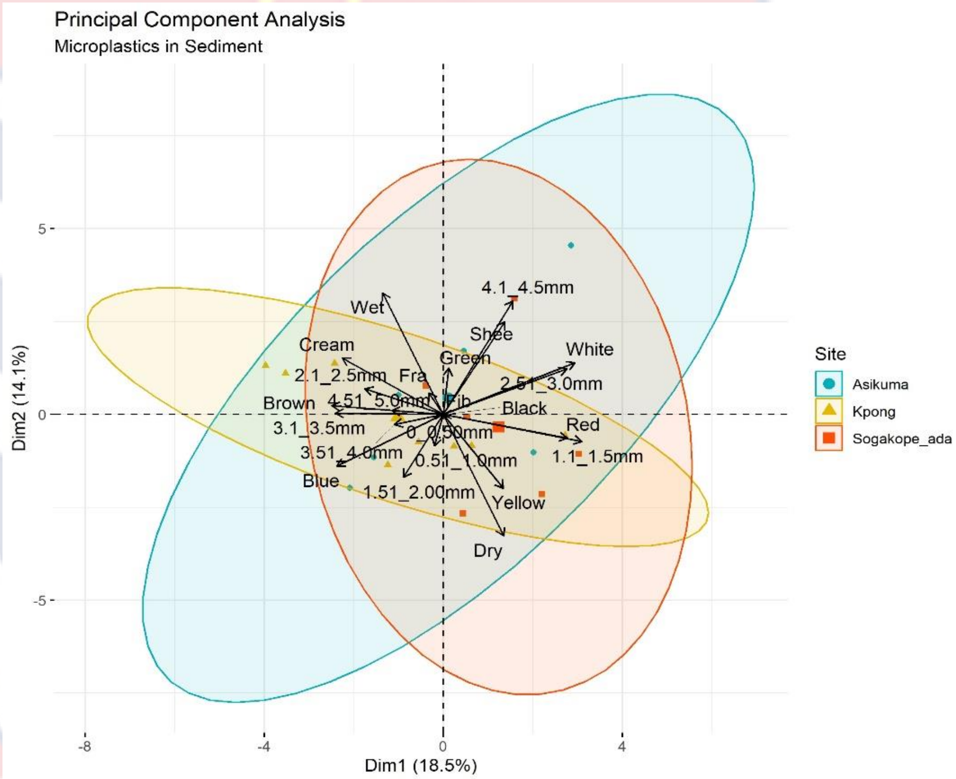

Figure 30: MPs variables in sediments from Asikuma Labolabo, Kpong and Sogakope Agodormi. (Dim: dimension/components).

With respect to sediment, (Fig 30 above) Dimension 1 was characterized by MPs sampled in the dry season. These MPs were in a small size class of 1.10-1.50 mm, medium size class of 2.51-3.00 mm and large size class of 4.10-4.50

mm. MPs here were sampled from all three sampling sites. A second group in this dimension are blue-coloured MPs of small size class of 1.51-2.00 mm and medium size class of 3.1-3.5 mm and 3.51-4.00 mm sampled from Kpong and Asikuma Labolabo. Dimension 2 consisted of MPs sampled in the wet season, from all three sites. MPs were characterized by colours such as cream, brown and blue and in medium size class of 2.10-2.50 mm and large size class of 4.51-5.00mm. From dimension 1 and 2, MPs in sediment were influenced by all size classes.

Estimated Exposure Assessment of Microplastics

Microplastics are unintentionally consumed by humans through food derived from a wide variety of sources, including fish. The consumption of MPs exposes humans to some health risks; thus, the need-to-know concentrations present in food/fish to take necessary precautions and minimize risks of exposure. The level of exposure can be determined using a simple parametric approach. This study primarily investigated the annual fish (contaminated with MPs) consumption for adult populations in Ghana. The estimated annual intake (EAI) of microplastics (through fish) was based on assumption that fish consumed were degutted; thus, excluding the gastrointestinal tract, using the equation:

$EAI = C \times AIR$ (Makhdoumi, Hossini, & Pirsaheb, 2023)

1) AIR is the annual ingestion rate of fish for Ghana (28 kg per capita) according to FAO (2016)
2) C is the average microplastic concentration based on fish muscle/edible tissue weight (ww)

$$C = Average\left(\frac{\text{number of MPs in edible tissue of fish}}{\text{weight of fish in grams}}\right)$$

Estimated MPs concentration for individual fishes were as follows: -

0.016 ± 0.017 items/gram (*Oreochromis niloticus*)

0.053 ± 0.054 items/gram (*Chrysichthys nigrodigitatus*)

0.065 ± 0.047 items/gram (*Chrysicthys auratus*)

0.055 ± 0.025 items/gram (*Tilapia zilli)*

0.016 ± 0.018 items/gram (*Tilapia guineensis)*

0.011± 0.001 items/gram (*Malapterurus electricus)*

9.00 ± 2.943 items/gram (*Brycinus brevis)*

0.093 ± 0.051 items/gram *(Parachanna obscura)*

0.030 ± 0.007 items/gram (*Auchenoglanis occidentalis)*

0.010 ± 0.008 items/gram (*Mormyrus* spp)

0.059 ± 0.059 items / gram (*Synodontis vellifer*)

As the current data on microplastic consumption via fish presents limited findings on human exposure, other food sources including freshwater fish species not studied here, poultry, beef, dairy, vegetables, seafood, bottled water and bottled drinks would provide a relative exposure risk associated with our Ghanaian estimates of microplastics in food. Other kinds of fish consumed from the marine environment have been excluded as the focus is on freshwater environs; thus, a drastic underestimation of Ghanaian freshwater fish from the Lower Volta Basin is presented in this thesis.

CHAPTER FIVE

DISCUSSIONS

Microplastics Ingested by Fish

From the results obtained, MPs were high in cultured species than wild species at each of the sites (Fig 2 above). This depicts that cultured species consumed more MPs which could come from the feed they consumed, coupled with those ingested from their environment. A study by Gündogdu, Eroldogan, Evliyaoglu, Turchini, Wu, (2021) recorded the prevalence of MPs in 25 out of 26 commercial fishmeal samples originating from 11 countries on four continents and Antarctica. Another study by Karbalaei *et al*. (2019) identified MP loads in three Malaysian commercial brands of fish meal investigated. With regards to the prevalence of MPs in aquaculture environment, Chen, Li, and Wang, (2021) stated that the concentration of microplastics in aquaculture environments is higher than in surrounding environments because of expanding aquaculture activities. Expanding activities can contribute high concentrations of secondary MPs from plastic aquaculture inputs such as nets and ropes. They further reported that fish or aquatic organisms may confuse MPs as prey and ingest them as food. Furthermore, Elizalde-velázquez and Gómez-oliván (2021) stated that the ingestion and concentration of microplastics in an organism can be influenced by the availability and concentration of MPs in the environment. The type of MPs identified in an organism can reflect the type of MPs present in the environment.

For this study, fibres were the most abundant MP shape identified in both cultured and wild fish species (Fig 7, 8 above). These fibres could originate as secondary MPs undergoing biodegradation from nets, ropes and other aquaculture inputs, as well as activities from terrestrial environments. Sultana *et al.* (2023) reported fibres to be the bulk of MP shape identified in both farmed fish collected from Mymensingh and Chandpur fish markets, and wild fish from the Padma River, Turag River, and Bhairab River in Bangladesh. They reported that most of the MPs originated from terrestrial environments (Sultana *et al.*, 2023). Pappoe *et al.* (2022) in their study, also reported fibres to be the primary MP shape observed in four marine species obtained from Jamestown fish landing beach in Ghana. They stated that the fibres could be linked to discharges from abandoned, lost, or otherwise discarded fishing gear emanating from trawl net and ropes which has been identified as a substantial source of marine litter worldwide. Another study by Silva-Cavalcanti, Silva, França, Araújo and Gusmão, (2017) reported fibres as the dominant MP shape consumed by *Hoplosternum littorale*, a common freshwater fish heavily consumed by humans in semi-arid regions of South America. They also reported/suggested urbanization to be the source of MPs in the river, which are eventually consumed by the fish.

With respect to the two tissues analyzed, there was higher abundance of MPs in gills (cultured- 380.67 ± 112.22, wild- 92.00 ± 67.88) than guts (cultured- 340.33 ± 147.55, wild-76.67 ± 58.23) for both cultured and wild species at all sites (Fig 4-6 above). The higher MP abundance in the gills could be due to the efficiency of the filtration apparatus of the gills (high filtration area and small gill raker spacing) and passive uptake of MPs during swimming and respiration. A study by Feng *et al.* (2019) reported a higher abundance of MPs in gills than gut for *T. kammalensis,* which was one of the six major wild fish from an important fish farm and mariculture area in Haizhou Bay, China. The higher prevalence in the gills of the fish was attributed to its mode of feeding -filter feeder and its place of habitat-in estuary where MPs are usually abundant (Feng *et al.*, 2019). Another study by Hosseinpour, Chamani, Mirzaei and Mohebbi-nozar, (2021) reported that there were higher MPs abundance in gills than guts of 14 fish species from the Persian Gulf. Parker, Andreou, Green and Britton, (2021) also compared and reported the abundance of MPs in the gills of pelagic fishes to be higher than that of the demersal. Even though the study did not state the possible reasons for the observation, it could be attributed to the efficiency of the filtration apparatus/gills or passive uptake of MPs during swimming and feeding (Parker *et al.*).

In comparing MPs abundance in fish at the three sampling sites (Asikuma Labolabo- 31.55 ± 20.55, Kpong- 22.23 ± 14.44, Sogakope Agodormi- 13.73 ± 7.81) Asikuma Labolabo reported the highest MPs (Fig 2, 3 above). The high MP abundance at this site could be from activities of humans on the bank of the river and other external inputs of fishing activities, as well as MPs from aquaculture installations in the waterbody. Pappoe *et al*. (2022) stated that laundry and domestic discharges have been reported to be responsible for considerable fraction of MPs (fibres) in aquatic biota. Yin *et al*. (2019), in their work, stated that the intensity of human activity is related to a higher level of microplastic pollution. Eerkes-medrano and Thompson (2018) stated that particles and fibrous (MPs) are released into aquatic environment from wastewater after washing of clothes/laundry activities. Chen, Li, and Wang, (2021) stated that rivers provide a freshwater aquaculture environment and are also considered a pathway for MPs from land to the marine environments. They also mentioned that in aquaculture environments, plastic fishing gear is the predominant and most important source of microplastics (Chen *et al.*). Plastic products such as fishing nets, fishing ropes, and floating balls are used in offshore cages and raft culture. Their aging and damage cause a large number of plastic fragments to enter the aquatic environments.

Black, blue and red being the frequent coloured MPs (fibres) recorded in this study corresponds to the colours of nets, ropes, floats etc. of aquaculture installations and colours of MPs from the external environment which may be attractive and confusing for organisms as prey (Addo *et al.*, 2022).The black MPs appear as prey to aquatic organism; thus, they feed on it as food *(Addo et al.*, 2022). Barboza *et al*. (2020) who recorded blue as the most abundant colour stated that blue MPs have a high probability of being up-taken by fish than MPs of other colours, as colour is an important clue for prey for some type of predators, and may be ingested by mistaking it for a prey (Barboza *et al.*, 2020). Kumar, Ravikumar, and Jeyasanta, (2018) recorded black, red and translucent fibre MPs in two species of fish bought from Thirespuram and Punnakayal fish landing sites at Tuticorin, India.

Most of the MP sizes recorded in both cultured and wild species were less than 2 mm (87 %) (Fig 9 above). This size range is smaller than what was recorded in the sediment (Fig 17 above) and water (Fig 21 above) but however, confirms with the majority of MP sizes recorded in the feed (66.67 %) (Fig 24 above). Such small-sized microplastics are considered to pose the most serious potential threats to both aquatic organisms and ecosystems (He, Goonetilleke, Ayoko and Rintoul, 2020). The small size of microplastic particles and their appealing coloration and buoyancy allows for easy picking and ingestion by fish

(Parvin, Jannat, & Tareq, 2021). In studies of McNeish *et al.* (2018), MP fibres were classified as small (< 1.5 mm), medium (1.6-3.2 mm) or large (> 3.3 mm); small fibres (MPs) were the most common size found across water and fish samples. Feng *et al.* (2019) reported that MP size < 1000 µm accounted for more than two-thirds of the total MPs in fish samples. They stated that compared to larger MPs, smaller MPs with the same mass can carry more toxic matter and is more difficult to remove as well.

In this study, the overall MP abundance recorded in fish, in the wet season (264.33 ± 26.44) was higher than the dry season (222.80 ± 75.66). This could result from urban run-off and stormwater that influences the temporal pattern or concentration of MPs in aquatic environments (Singh *et al.*, 2021).

Spatial Distribution of Microplastics in Water and Sediment

The results from the analysis indicated that fibres were the most dominant MPs (as recorded in the fish) in both water (fibres- 36.67 ± 14.57, fragments- 2.33 ± 0.58) and sediment (fibres- 322.00 ± 144.44, fragments- 2.67 ± 3.44, sheets- 0.33 ± 0.82) at all 3 sites and both seasons. (Fig 15, 19 above). These fibres as discussed earlier may come from land-based human activities such as wastewater from laundry/washing of clothes (thousands of fibres may be released per wash), fishery and aquaculture activities, as well as atmospheric deposition (Chen *et al.*, 2021). Furthermore, the distribution of MP fibres in water and sediment could mean that MPs have different properties or characteristic that enable them to float or sink. In studies of Liu *et al.* (2021) to find the distribution of MPs in water, sediment and fish of the Dafeng River, a remote river in China, recorded fibres to be the abundant MPs in fish, water and sediment. They stated that MPs, being anthropogenic in origin, footprints of which are in the environments, are determined by anthropogenic influences. They identified that the shape, characteristics, and polymer composition have been suggested as links for source identification (Liu *et al.*, 2021). Another study by Vermaire, Pomeroy, Herczegh, Haggart and Murphy, (2017) observed microfibres as the dominant MP shape in both water and sediment from the Ottawa River in Canada. They stated that microfibers come from multiple sources and are transported into aquatic system both by runoff and by atmospheric deposition. In comparing MPs abundance in water and sediment of the three-sampling point with reference to the wet season (water- 37.00 ± 14.93, sediment- 91.2 ± 9.45) (Fig 12, 18 above), an 8-fold increase of MPs in sediment was observed. This could mean that most of the MPs have characteristics such as high density that enables them to sink to the bottom of the waterbody or settle in sediments. On the other hand, MPs in the water column combine with organic or inorganic substance that make them heavy or dense and sink into the sediment. In studies of Liu *et al.* (2021), the abundance of MPs was reported to be four orders of magnitude higher than in surface water. Sediments were/are considered to be reservoirs of MPs, and the level of pollution in sediment has been reported to be several orders of magnitude higher than in surface water (Liu *et al.*, 2021). Blankson, Tetteh, Oppong and Id, (2022) concluded that MP abundance in sediment was higher than water, as MPs are associated with sediment rather than water. This increases the potential for MP accumulation in filter-feeders and soil-dwelling biota compared to pelagic species. In a review, Wu, Xiong, Hossein, Zhang and Xu, (2022) stated that MPs are about one or two orders of magnitude higher in sediment than in water, pointing to sediment as an important sink for MPs in aquaculture systems.

In reference to water and sediment (Asikuma Labolabo- 165.5 ± 122.5, Kpong- 78.5 ± 58.5 and Sogakope Agodormi- 267.50 ± 219.50) (Fig 12, 13, 18 above), MPs abundance was highest at Sogakope Agodormi. This could be due to the high number of cages and aquaculture activities at that site compared to the others. Sogakope Agodormi is a major aquaculture production area situated close to a potable water treatment plant. Although Kpong University farm is equally an aquaculture area, the number of cages identified were less, with less human activities compared to Sogakope Agodormi. Asikuma Labolabo, is within stratum 2 of the Volta Lake where fish farming is intensive (Kassam, 2014). However, the abundance of MPs was lesser than expected. This could be due to the currents and direction of water, carrying most of the MPs away from the area. Kim, Seung-kyu, Kim, Choi and Woo (2015) stated that seasonal winds and currents could be considered possible reasons for dramatically lower microplastic abundance on one of the beaches in their studies. Secondly, activities from the potable water treatment plant cannot be ignored. Wastewater from the plant may be discharged into the waterbody and can be a source of MP pollution in the water. Microplastic contributions from cages and treatment plants can increase the abundance of MPs in the waterbody at this site (in both water and sediment). Chen *et al.* (2021) stated that the concentration of MPs in aquaculture environments is generally higher than in surrounding environments because of rapidly expanding aquaculture activities. Microplastics can be released into aquaculture environments from plastic fishing gear as well as fish feed and medicine. Wu *et al.* (2022) also reported that compared to other freshwater environments, MP pollution in freshwater aquaculture systems is high; this may be related to their relatively closed environments (Wu *et al.*, 2022). A study by Sun, Dai, Wang, Van Loosdrecht and Ni, (2019) indicated that wastewater from treatment plants (WWTPs) play an important role in contributing MPs to natural aquatic systems. Chen *et al.* (2021) also stated that industrial, agricultural and domestic wastewater contains microplastics which are also major contributors of microplastics in aquatic environments.

Frequent coloured MPs such as black, red and blue recorded in the fish were extracted from sediment and water. This could suggest that MPs in the fish, as well as sediment and water could be of similar source/origin. These sources could include cage nets and or materials which were/are being used at various aquaculture farms sampled (as the colours of the MPs correspond with the colours of these cage materials). Other coloured MPs such as cream, brown, yellow, green, brown etc. could originate from external sources such as synthetic textiles, hygiene and cosmetic products and the fishing industry. Singh *et al.* (2021) reported transparent, white, black, green, yellow, and blue as colours of MPs in water and sediment of the Dafeng River in China. Peng, Xu, Zhu, Bai and Li, (2018) also reported transparent, red, black, blue and white as colours of MPs identified in sediment of six rivers and one tidal flat in Shanghai urban districts of China. They stated that the rivers were a hotspot for microplastic pollution which came from human activities (Peng *et al.*).

Most of the MPs in water measured between 1.50-5.00 mm whilst majority of those in the sediment measured between 0.50-3.00 mm. Thus, MPs in water were relatively larger than in sediment. The relatively large but less dense nature of the MPs in water increases the potential of it being transported by water (by the currents and waves). Singh *et al.* (2021) reported that MPs in water and sediment of the lower Ganga River were categorized into size classes of 2.50-5.00 mm and > 5.00 mm. The latter had the maximum number of MPs in water samples while size classes of 2.50-5.00 mm contributed a greater number of MPs in sediment samples. They stated that the varying size of MPs play a crucial role in their distribution and transport in the environment, and influences their bioavailability (Singh *et al.*, 2021). Blankson *et al.* (2022) also reported that the difference in the mean size of MPs between water 1.21 ± 0.26 and sediment 0.27 ± 0.05 was about 4.5-fold increase for the Weija Dam and the Densu Delta. The mean size of the sediment was 0.84 ± 0.13, and that for water was 1.01 ± 0.11. They observed that the stagnant water system of the Dam promoted floatation of larger-sized microplastics while the flowing waters of the Delta did not show any selectivity in the deposition of MPs between sediment and the water column (Blankson *et al.*).

Presence and Abundance of Microplastics in Feed

The results from analyses indicated that MPs abundance in Feed A- 25.0 ± 33.23 items/g was higher than Feed B- 15.67 ± 15.06 items/g and Feed C- 6.67 ± 9.43 items/g. Thus, MPs abundance was in the order of Feed A > Feed B > Feed C (Fig 22 above). The differences in abundance of MPs between the feed could be as a result of the part/component of the fish used in the feed formulation. Some manufacturers use the whole fish while others use by-products such as the viscera, head, and gills. Fish brands that use the whole fish are likely to contain less MPs than those formulated with by-products. Secondly the type of fish used in manufacturing fish meal, be it a pelagic or demersal can influence the quantity of MP contamination in the feed. Demersal fishes might contain more MPs than pelagic fishes as more MPs settle in sediments. In studies of Karbalaei *et al.* (2019), analyses of three feed brands A, B and C, with. brands (A and B) mainly made up of whole fish (mixture of flesh and viscera), and brand C from fish waste (gastrointestinal tract, scales, fish head and bone) produced different microplastic abundance. From their result, they observed that Brand C contained significantly higher MPs compared to A and B. A study by Gündogdu, Eroldogan, Evliyaoglu, Turchini and Wu, (2021) analysed 26 different fishmeal products, originating from 11 countries on four continents and Antarctica but did not specify the part of fish used in the fish feed. From their analysis they observed that fishmeal obtained from China and Morocco had relatively higher MP content, whereas no microplastics was detected in krill meal (a type of fish feed) obtained from Antarctica. They stated that differences and heterogeneity in microplastic concentrations could likely be due to differences in the contamination level of the actual species used for fishmeal production (Gündoğdu *et al.*). Different feeding modes can significantly influence the microplastic content of organisms and thus, the contamination level of the fishmeal derived from those species.

The dominant type of MPs isolated in all three feed was fibres (42.67 ± 26.63), followed by fragments (4.33 ± 5.86) and sheets (0.33 ± 0.58) (Fig 23 above). This is in agreement with the primary morphology of MPs found in the guts and gills of cultured species. This finding is in agreement with a study by Castelvetro *et al.* (2020) who also reported microfibres as the dominant MPs isolated from fish meal samples. The microfibres were reported to have typically originated from synthetic textiles. A review by Mahamud *et al.* (2022)

reported that microfibres were the highest- 80 % documented occurrence of MPs in fish meal studies. They stated that MPs are found in almost all marine habitats and are about the same size as sediments and some planktonic species, making them bioavailable to various aquatic organisms, including fish.

Most of the MP sizes recorded in the feed were < 2 mm (66.67 %). This corresponds to the majority of size recorded in the fish, which could also imply that some of the MPs ingested by the fish could come from the feed. Mahamud *et al.* (2022) again reported that MP size obtained in fishmeal ranged between 1 and 3 mm; this corresponds to sizes of MPs in the stomach content of neotropical omnivore fish species. Another study by Way, Hudson, Williams, Langley and Marsh, (2022) who compared newly developed methods of extracting MPs from fish meal recovered MPs in the size range of 0.25-1.00 mm.

Black, red and blue were the most abundant coloured MPs recorded in the fish and other environmental samples, and also in fish feed (Fig 24 above). This could imply that some of the MPs recorded in fish came from the feed. These coloured MPs (black, red, and blue) as well as others recorded (yellow, violet, orange and white) as mentioned above, could come from MP-contaminated fish and or packaging materials of other raw materials/food stuffs used for the feed formulation. Gündogdu *et al.* (2021) recorded black, blue, brown, cream, orange, pink, purple, red, yellow etc., as some colours of MPs obtained from fish meal. They stated that the main source of the plastic content of aquatic products is the plastic pollution in the marine environment. However, conditions during processing and packaging of fishmeal may also contribute a certain level of plastic pollution in fishmeal, which can enter the fishmeal products at many stages during the processing.

Overall MP Abundance at the Three Sampling Sites

The overall MPs in all matrices analyzed- fish, water, and sediment- was highest at Asikuma Labolabo (Fig 24 above). This was influenced by the high MP abundance in fish (both cultured and wild- 631 MPs; mean of 315.5 ± 273.65) compared to 445 MPs (mean- 222.25 ± 144.60) and 288 MPs (mean- 143.75 ± 166.52) for Kpong and Sogakope Agodormi, respectively. The high MPs abundance in fish at Asikuma Labolabo means that the fish in that area ingested more MPs due to increased exposure to higher MP concentrations. The higher MP concentration in both cultured and wild species at Asikuma Labolabo could come from activities of human settlement close to the bank of the river. Secondly run-offs from the land during rainy periods could carry a lot of debris including MPs which could contribute to its abundance in the water. Furthermore, the flow/velocity of the water close to the banks of the river, where the cages are mounted, is slow. Thus, the river may contain more suspended MPs which potentially get ingested by caged fish. Compared to downstream areas of Kpong and Sogakope Agodormi, human settlement and activities at the bank of the River contribute lesser MPs to the water, thus a limited amount/concentration is expected to float within the river around Sogakope Agodormi. Moreso, the velocity/flow of the water at these two sites is fast with less suspended MPs in the water; these transported suspended microplastics are potentially ingested by fish. Thus, human activities coupled with the hydrology of the river influenced/contributed to the high abundance of MPs in fish at Asikuma Labolabo.

Principal Component Analysis for Fish samples

With respect to the distribution of MPs in fish, a study by Rasta, Sattari, Taleshi and Nanim, (2021) reported black, red and blue as the most frequent colours in different tissues of some commercial fish species from Anzali Wetland in the Southwest Caspian Sea, Northern Iran. Fibrous MPs were more in fishes from these sites because of the high activities of cage farming in these areas (Rasta *et al.*, 2021). A study by Kasamesiri and Thaimuangpho (2020) reported fibres as the major type of MPs found in fishes in the Chi river in Thailand. They stated that the fibres came from the degradation of fishing gear, fish cages or nylon ropes and sewage from washing clothes.

On the lower Volta Lake, cage farming is dominant in Stratum 2 within the Asuogyaman district and surrounding communities including Asikuma and Kpong (Kassam, 2014). *Tilapia zilli*- a demersal wild species- sampled from Kpong contributed blue and red coloured MPs. The sizes of MPs consumed by wild species at Kpong ranged between 1.51 and 2.00 mm, and 4.51 and 5.00 mm. Mcneish et al. (2018) classified MPs in size classes of small (<1.50 mm), medium (1.60–3.20 mm), or large (>3.30 mm), and reported the presence of the size classes in fishes analysed. Rasta *et al*. (2021) stated that organisms (fish) may ingest MPs which have similar sizes to the food they consume. Some fish food include plankton, polychaete, copepod (Borges-Ramírez, Mendoza-Franco, Escalona-Segura and Osten, 2020). Hastuti, Lumbanbatu and Wardiatno, (2019) stated that omnivorous fishes often ingested fibres, while fragments were mostly ingested by benthic organisms. This was attributed to the fact that fibres had lower size and density due to its polymer composition. MP polymeric fibres are mostly low-density polyethylene (PE) and polypropylene (PP) (Rasta *et al.*, 2021). These fibrous MPs could come from cage materials, as well as synthetic fibres from terrestrial activities which are washed into the waterbody (Hastuti *et al.*, 2019). Microplastic sizes ingested by the species ranged from small size of 1.51-2.00 mm, through medium size of 2.10-2.50 mm to large size of 4.51-5.00. mm. These could be explained by the fact that these sizes corresponded to the sizes of food they consume. Rasta *et al.* (2021) stated that fishes may ingest MPs with the same size as their food.

Principal Component Analysis for Water samples

Microplastics distribution in water at all three sites were mostly medium to large size classes. Although relatively large, MPs are able to float in the water column because of the low-density polymers they are produced from. These polymers mostly include polypropylene (PP), polyvinyl alcohol (PVA), polystyrene (PS), low-density polyethylene (LDPE), polyamide (PA) acrylic etc. Li, Busquets & Campos, (2020) in a review reported MPs polymers that have been identified in freshwater systems to include polypropylene (PP), polyvinyl alcohol (PVA), polystyrene (PS), polyamide (PA), polyethylene terephthalate (PET), polyvinyl chloride (PVC), polyurethane (PU) and others. With aquaculture activities from all three sites, it could imply that most of these MPs were coming from the aquaculture installations as well as some external sources (Li *et al.*).

Principal Component Analysis for Sediment samples

With respect to MPs distribution, a review by Yang, Zhang, Kang, Wang & Wu, (2021) reported colours of MPs that have densified in sediment to include white, transparent, red, yellow, green, brown, gray, etc. They also stated that some study recorded MPs size in sediment to be in the range of 1 to 5 mm. The different colours and size classes of MPs identified in sediment may be due the different types of polymers in their composition. Most often than not, MPs of dense polymers tend to sink/settle in the sediment. Some of these dense polymers are polyester and rayon. Borges-Ramírez *et al.* (2020) reported acrylic, polyester, polyethylene terephthalate as some dense polymers identified in sediment in their study. MP abundance was almost the same for both seasons. From dimensions 1 and 2, MPs in sediment were influenced by all size classes. Furthermore, with aquaculture activities at all three sites, it could imply that MPs from the aquaculture installation contributed to MPs in the sediment. Thus, MPs abundance in the sediment could be influenced by the aquaculture activities in the waterbody.

Exposure of Microplastics to Humans

The estimated annual intake (EAI) of the various species in the study suggested that a person who consumes cultured *Oreochromis niloticus* could be exposed to 441.37 gram/person/year. The annual dietary exposure for Ghanaians consuming freshwater benthopelagics are listed as follows: *Tilapia guineensis* (434.53 gram/person/year), *Synodontis vellifer* (1647.69 gram/person/year), *Parachanna obscura* (2447.06 gram/person/year) and *Malapterurus electricus* (312.96 gram/person/year). The consumption of demersal freshwater fish could expose the Ghanaian population to MPs from species such as *Auchenoglanis occidentalis* (828.23 gram/person/year), *Brycinus brevis* (2595.46 gram/person/year), *Chrysichthys nigrodigitatus* (1483.66 gram/person/year), *Chrysichthys auratus* (1824.55 gram/person/year), *Mormyrus spp*. (287.10 gram/person/year), and *Tilapia zillii* (1541.34 gram/person/year). From the estimation, most of the demersals had a higher exposure annual intake than the cultured and wild benthopelagic species. This could be attributed to their habitation- at the bottom of the water column, close to the surface of bottom landing. They feed from the sediment, and the bottom of the water column (Borges-Ramírez *et al.*, 2020). Thus, their habitat exposes them to high levels of MPs. Studies by Makhdoumi *et al.* (2023)- 232,260 to 8,864,100 P/kg/bw/year, Pappoe et al. (2022)- 1307 to 29647 microplastic particles per year, and Addo *et al.*(2022)- 2,600 MP items/year reported high MP estimated intakes in fish assessed although, most of these studies were in the marine environment. This study therefore provides baseline data of estimated annual intake of MPs for some freshwater fish species in the Lower Volta Basin in Ghana. The EAI observed for the fish in this study were within the range of 518 to 3078 MP items/year/capita which is the recommended EAI for some selected European and American countries (Makhdoumi *et al.*, 2023). The presence of accumulated microplastics in fish may not only reflect contamination in freshwater bodies such as rivers, lakes, stream etc. but also mirror the potential dangers to human health posed by the ingestion of fish contaminated with microplastics (Pappoe *et al.*, 2022). The subtle effect of plastic ingestion on human is, however, not well understood but there is documented evidence of physical damage to sensitive organs via plastic ingestion (Addo *et al.*, 2022). The toxicities of microplastic ingestion, however, could be manifested by interfering with human body functions such as the digestive system (Zhang *et al.*, 2021), cardiovascular system (Bhuyan, 2022), endocrine system (Prata, Costa, Lopes, Armando, & Rocha-Santos, 2019), the nervous system (Rahman, Sarkar, Yadav, Achari & Slobodnik, 2021) and reproductive system (Ragusa *et al.*, 2021); thus, the need to take measures to minimize exposures.

Health Implications of MPs in Humans

MPs are ubiquitous environmental contaminants which pose risks to human health; it has been shown that they can be ingested via a wide range of aquatic organisms, both freshwater and marine, and can therefore be accumulated via the food web (Sana, Dogiparthi, Gangadhar, Chakravorty & Abhishek, 2020). Exposure of humans to MPs may occur via consumption or ingestion, inhalation, and skin contact through food, water, air, and consumer/packaging products (Bhuyan, 2022; Rahman *et al.*, 2021). Health related issues such as oxidative stress, cytotoxicity, neurotoxicity, immune system disruption, and transfer of MPs to other tissues have been reported with MP contamination in the body (Bhuyan, 2022). The ingestion of food and water contaminated with MPs is the primary route of human exposure. Food products contaminated with MPs, are frequently consumed by humans, including bivalves, crustaceans and commercial fish, sugar, salt, beer, honey and bottled water (Rahman *et al.*, 2021). Most often than not, aquatic organisms such as shellfish- clams and mussels- and a few other shrimps, known to contain high concentrations of MPs, are consumed wholly with their intestines and gills

(Sana *et al.*, 2020). Vázquez-rowe, Ita-nagy and Kahhat, (2021) reported that high concentrations of MPs consumed increase human exposure. It has been documented that exposure of Europeans through the consumption of bivalves has been estimated to be 11,000 microplastics person^{-1} year^{-1}, while that of table salt has been calculated as 37 microplastics person^{-1} year^{-1} (Prata *et al.*, 2020).

Inhalation is an involuntary action of breathing in air. The air may contain MPs released from numerous sources, including synthetic textiles, abrasion of materials (e.g. car tires, buildings) and resuspension of microplastics on surfaces, waste incineration, and landfills (Rahman *et al.*, 2021). As such people working at synthetic textile mills and landfills may be at a high risk of exposure. In agreement with this, Rahman *et al.*(2020) reported that synthetic textile mill workers with occupational exposure to airborne MPs were associated with severe respiratory symptoms due to the development of inflammatory airway and interstitial lung diseases. Also, MP contamination through the skin may occur through contaminated air (dust), water, and products like skincare, and washing products (Sana *et al.*, 2020). The material that enters the human body occurs as MPs and microbeads, and dominantly as nanoplastics (Sana *et al.*, 2020). From the above-mentioned sources, microplastics are absorbed via skin, the mucous membrane, and gastrointestinal mucosa. Some MP polymers that have been identified in the human body include Polyethylene (PE), Polystyrene (PS), Polypropylene (PP) (Hwang, Choi, Han, Choi & Hong, 2019).

Plastic polymers are defined as non-toxic because they are not reactive and generally cannot be easily transported across biological membranes due to their size. However, non-polymeric substances, like chemical additives such as plasticizers, flame retardants, pigments, antimicrobial agents, heat stabilizers, UV stabilizers, fillers, and flame retardants such as polybrominated diphenyl ethers (PBDEs) or residual monomers can be hazardous to human health and the environment when they leach from the plastic polymer matrix (Smith, Love, Rochman & Neff, 2018). These chemical additives can accumulate persistent organic pollutants (POPs) in MPs which could be hazardous to the health of aquatic animals, and subsequently humans.

Oxidative stress occurs when MPs release oxidizing chemicals (such as metals) which are adsorbed to their surfaces and reactive oxygen radicals from the host body that are produced during inflammatory response (Prata *et al.*, 2020). Oxidative stress due to the exposure to MPs was reported in mice and zebrafish as well as humans. In humans, limb and joint prostheses containing MPs were reported to release acute toxicants and free radicals due to an acute inflammatory response (Blackburn & Green, 2022; Prata *et al.*, 2020; Rahman *et al.*, 2021). Furthermore, MPs are found to be cytotoxic as an effect of oxidative damage and inflammation. Cytotoxicity happens as a result of some specific cells namely macrophages (an acute response cell of the body) internalizing MPs, and as the MPs are not membrane bound, they interact easily with the intracellular organelles, potentially causing injury. In vitro study reported by Prata *et al.* (2020), Rahman *et al.* (2021), and Schirinzi *et al*. (2017) showed that MPs at the level of exposure of 0.05-10 mg per liter increased reactive oxygen species to high concentrations, which contributed to cytotoxicity in human cerebral and epithelial cells. Human immunological function can be potentially altered by MPs resulting in autoimmune disorders. This happens as a result of chronic damage in cells, the production of immune modulators, and the incorrect stimulation of immune cells. All the antibodies against self-antigens could be produced as an outcome of the chain of events (Bhuyan, 2022). After exposure, MPs may translocate to distal tissues through the circulatory system. In the circulatory system, MPs have been reported to cause a systemic inflammatory response, blood cell cytotoxicity through internalization, vascular inflammation, occlusions and pulmonary hypertension (Rahman *et al.*, 2021). Translocation is especially likely during inflammation, due to the increased permeability of epithelial barriers. Prata *et al*, (2019) reported that MPs were translocated to organs such as the liver, spleen and kidney responsible for the metabolism and excretion of xenobiotics through the circulatory system. Other effects of translocation of MPs are the increased activity of osteoclasts due to bone loss caused by PE and PS particles (Prata *et al.*, 2020).

CHAPTER SIX

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

Summary

The Lower Volta Basin serves as a source of water for domestic use, agriculture purpose, transport of goods and services, inland fishing, and commercial aquaculture production. Lake Volta (which forms part of the Lower Volta Basin) hosts about 143 fish species and provides about 90 % of the total inland fish catch (Rurangwa, Agyakwah, Boon & Bolman, 2015). Moreso, 80 % of the total aquaculture production in Ghana is from the Lake (Asmah, Karikari, Abban & Ofori, 2014; Ministry of Fisheries and Aquaculture Development, 2012). On the Volta Lake, cages are the dominant culture fish production system. Most of these cages have components such as galvanized pipe frames, nets, ropes, lines and buoys/floats, drums, etc. made from plastics. These plastics can degrade or by biofouling, break-off with a resultant release of secondary microplastics (MPs) into the aquatic environment. The size of microplastic particles and appealing coloration allow for easy ingestion by fish. Microplastics efficiently adsorb persistent, bioaccumulative and toxic substances (PBTs) from the environment, and when ingested by fish, disturb their physiological functions, which then go through the food web creating adverse health issues in human.

Despite the recent surge in microplastic research in Ghana, there is limited information on MPs in the Lower Volta Basin. Thus, the study aimed at filling in knowledge gaps by providing baseline information on MPs sampled in cultured (*Oreochromis niloticus*) and wild fish species (*Chrysichthys* *nigrodigitatus*, *Chrysicthys auratus*, *Tilapia guineensis*, *Tilapia zilli*, *Mormyrus* spp, *Synodontis vellifer*, *Synodontis schall*, *Parachanna obscura*, *Auchenoglanis occidentalis*, *Brycinus brevis* and *Malapterurus electricus*), river water and sediment from Asikuma Labolabo, Kpong, and Sogakope Agodormi along the Lower Volta Basin, during the wet and dry seasons. The study also accessed the exposure to humans who consume MPs-contaminated fish. Three commercial fish feed used at the various cage-culture farms sampled were also analysed.

Microplastics were extracted from the gills and guts of both cultured and wild fishes by chemically digesting the two tissues. Water and sediment were density-separated and filtered whilst the fish feed was chemically digested. There was MPs present in all sample matrices.

In comparing MPs abundance in fish from the sites, the overall percentage abundance of MPs in both cultured and wild sites, for both seasons, was highest in Asikuma Labolabo (47 %), followed by Kpong (33 %), and Sogakope Agodormi (20 %); there were significant differences in MP abundance obtained from the three sampling sites/locations ($p < 0.05$). For sediment, Sogakope Agodormi recorded a high percentage MP abundance of 49 % followed by Asikuma Labolabo (34 %), and Kpong (17 %) for dry and wet seasons. There were no significant differences in MP abundance in the sediment at the three sampling sites ($p > 0.05$). For water samples for the wet season, Sogakope Agodormi (48 MPs, mean- 16 ± 19.05 items/L) > Asikuma Labolabo (43 MPs, mean- 14.33 ± 8.50 items/L) > Kpong (20 MPs, mean- 5 ± 0.82 items/L). There were no significant differences in MP abundance in water at the three sampling sites ($p > 0.05$). For the commercial feed, Feed A had 75 MPs-mean of 25 ± 40.71items /g. This was followed by Feed B (47 MPs -mean of 15.67 ± 18.45 items/g). Feed C had the least number of microplastics (20 MPs mean 6.67 ± 11.55 items/g); The difference in abundance was not significant among the three feed ($T = 0$, $p = 1.00$). Results from the estimated annual intake also indicated that Ghanaian adults who consume freshwater fish from the lower Volta basin are exposed to MP concentrations between 287.10 and 2595.46 MP gram person^{-1} year^{-1}.

Conclusions

The results showed that there was widespread microplastics in the Lower Volta Basin. Microplastic abundance was high in cultured fish species, in comparison to wild fish species. The high abundance of MPs in cultured species, implied that the majority of cultured species were exposed to, and ingested more MPs that had degraded from the aquaculture system. Coupled with MPs from aquaculture installations, human activities including laundry wastewater disposed into the Volta River/lake, and improper waste management around the river banks within the communities influenced the concentration of MPs in the Lower Volta Basin. A high abundance of MPs was recorded in the sediment, as compared to water. This implies that sediments are the primary sink or accumulation zone for MPs in the Lower Volta Basin. From the three different brands of fish feed analyzed, microplastics were present. Fibres were the most dominant microplastic identified in sample matrices, in addition to lower percentages of sheets/films and fragments.

The estimated annual intake values of MPs in fish in this study were of a similar range to other EAI values computed for some marine species in previous studies in Ghana, America and European countries. However, there is a need to take measures to minimize MPs exposure in fish.

This is the first study to assess the occurrence and abundance of microplastics (MP) in cultured *(Oreochromis niloticus*), and wild fish species (*Chrysichthys nigrodigitatus*, *Chrysichthys auratus*, *Tilapia guineensis*, *Tilapia zilli*, *Mormyrus spp.*, *Synodontis vellifer*, *Synodontis schall*, *Parachanna obscura*, *Auchenoglanis occidentalis*, *Brycinus brevis*, and *Malapterurus electricus*), sediment and water, in the Lower Volta Basin in Ghana, for the dry and wet seasons.

With respect to the fish species analysed, this study provided baseline information on microplastics in *Chrysichthys auratus, Tilapia guineensis, Mormyrus spp., Synodontis vellifer, Auchenoglanis occidentalis, Brycinus brevis and Malapterurus electricus*; these local fish species are being investigated for microplastics for the first time in Ghana. This is because literature reviewed has shown that no research work has been completed and published on them.

Recommendations

1. Further studies could be completed along the different sections of the Volta Lake to ascertain MP occurrence, shapes and distribution.
2. Sampling should be completed across and along the river to obtain a better representation of MP occurrence and distribution in the Lower Volta Basin.

3. Further research could be carried out on edible tissues (muscle) of fish from both cultured and wild fish from the Volta Lake, for possible MPs occurrences.
4. Fish feed should be standardized to reduce MP contamination.
5. Cage materials such as nets, ropes and floats that are made of plastics need to be changed to biodegradable materials such as raffia and bamboo to reduce the introduction of plastics into waterbodies through aquaculture.
6. Periodic monitoring should be completed to inform trends and changes in spatial distribution of microplastics in the Lower Volta Basin.
7. Research should be on-going to aid with waste management to reduce the plastic inputs into waterbodies. As the ingestion and accumulation of microplastics from fish is of public health concern, with resultant health issues, it is importance to monitor our waterways to reduce the plastics released into them.

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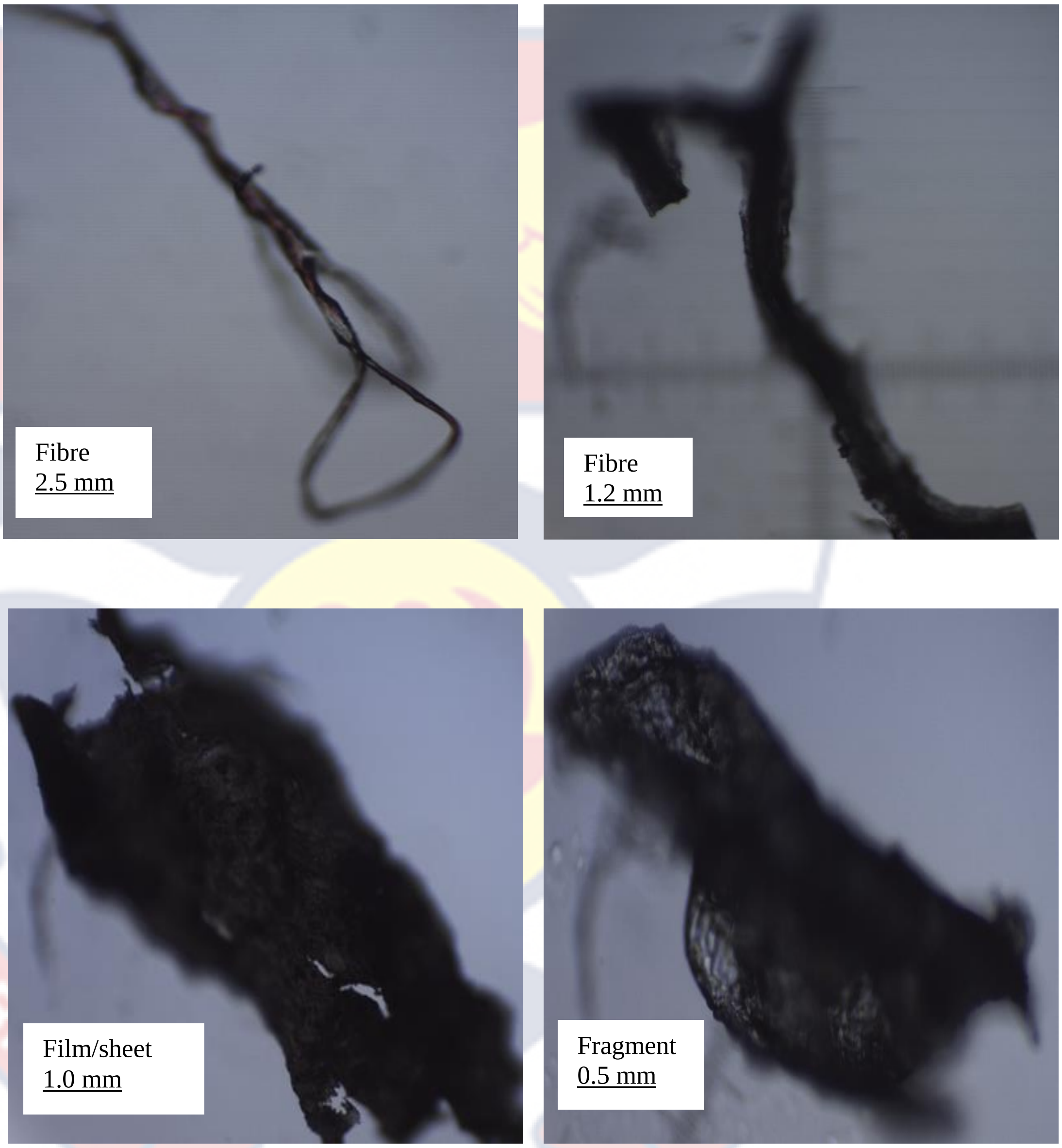
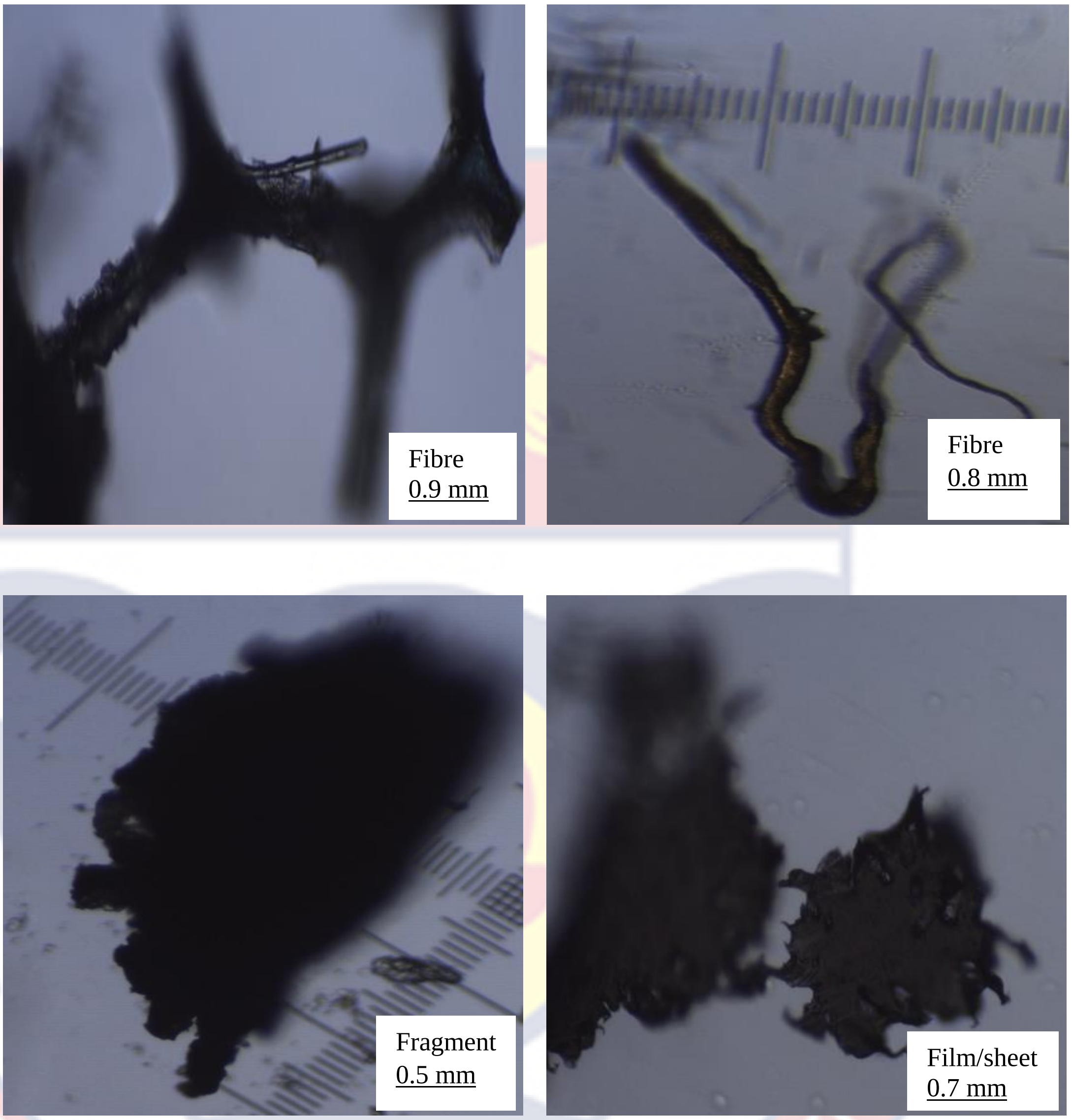
APPENDICES

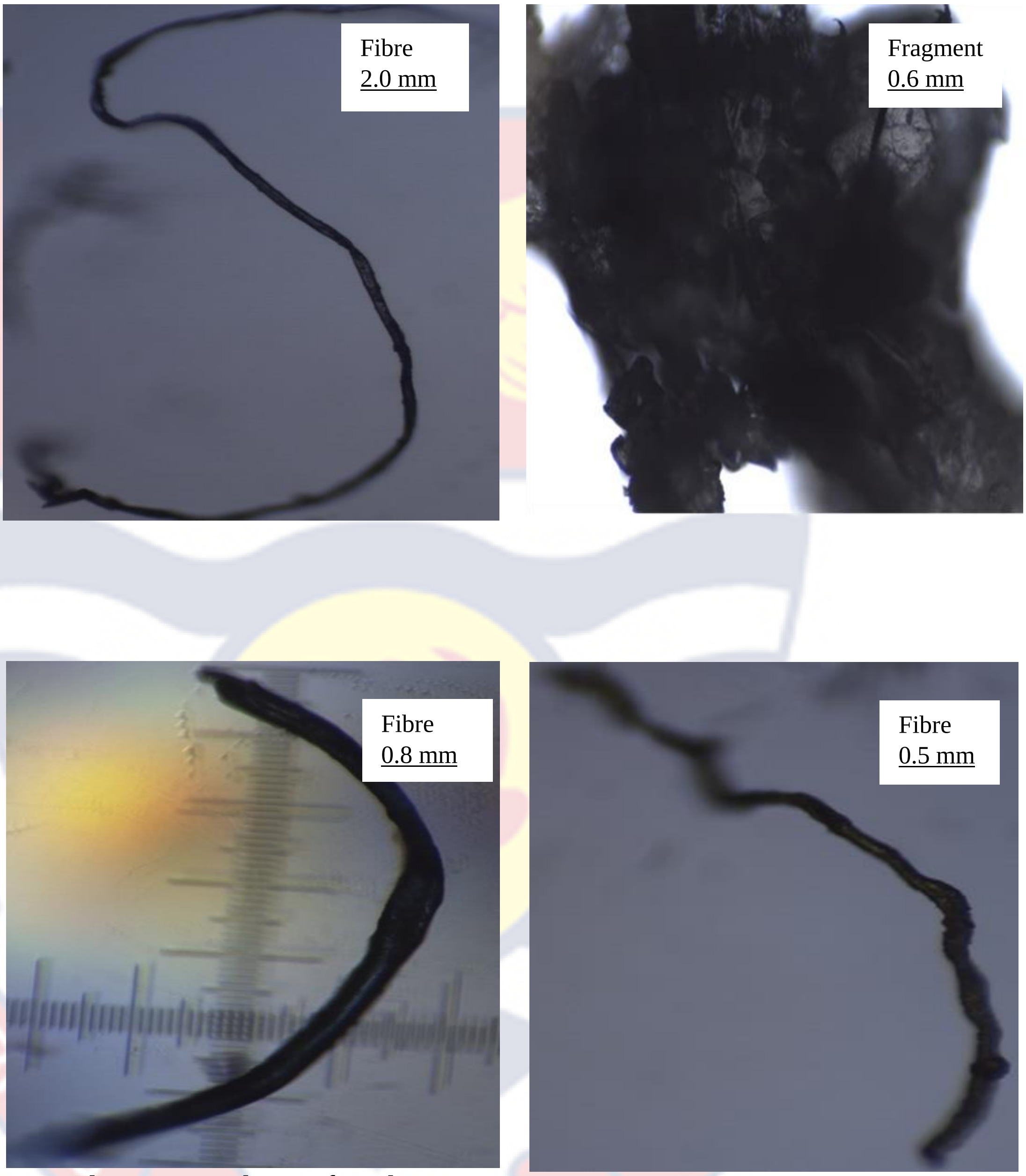
APPENDIX A: MICROPLASTICS PICKED FROM THE DIFFERENT SAMPLE TYPES



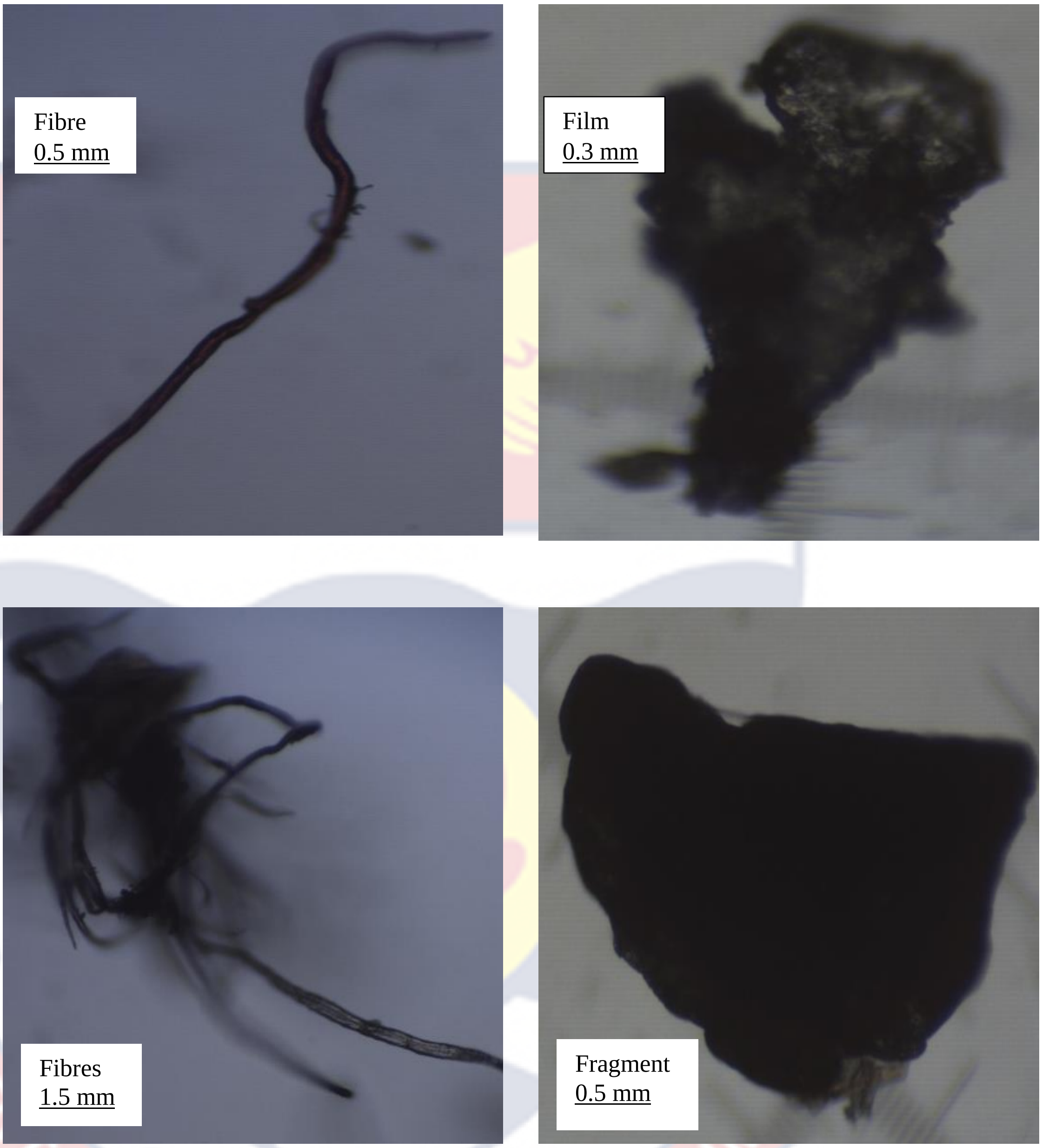
Appendix 1:Microplastics found in both cultured and wild fish species.



Appendix 2: Microplastics found in sediment.



Appendix 3: Microplastics found in water.



Appendix 4: Microplastics found in Fish Feed.

APPENDIX B: SAMPLES OF FISH SPECIES EXAMINED FOR PRESENCE OF MICROPLASTICS

Appendix 5 : *Oreochromis niloticus* (with gills and guts extracted).

Appendix 6: Mixed species -*Oreochromis niloticus, Chrysichthys nigrodigitatus, Synodontis spp.*

Appendix 7: *Tilapia guineenis* and *Mormyrus spp*

Appendix 8: *Brycinus brevis*

APPENDIX C: MICROPLASTICS STUDIES COMPLETED IN GHANA

Table 2: *A Compilation of Microplastic Studies Completed in Freshwater and Marine Environments in Ghana to Date (2020-2022).*

Author	Year (sampling)	Samples	Sampling sites	Size range	Polymer type	Results	Characterization	Microplastic type	Mean concentration	Total number of MPs detected
(Adika et al., 2020)	2019	fish	Tema fishing harbor.	0.5 – 4.75 mm	N/A	No identification of polymer types	Microscope	Fibre Fragment Sheet Bead pellet	*S. maderensis* (40.0 ± 3.8 items/fish), *D. angolensis* (32.0 ± 2.7 items/fish) *S. aurita* (25.7 ± 1.6 items/fish)	Not stated
(Gbogbo et al., 2020)	Not stated	water, sediment fish, crabs feacal matter of waterbirds	wetland of Sakumo II Lagoon	< 0.1 – 5 mm	N/A	No identification of polymer types	Microscope	Not stated	3 ± 2 items/fish 8 ± 1 items/crab 1.85 g^{-1} of sediment 0.09 ml^{-1} of water	128
(Chico-Ortiz et al., 2020)	2020	Sediment cores	Mukwei Lagoon Kpeshie Lagoon Mangrove forest	Kpeshie Lagoon: (26.63 ±5.88) µm Mangrove forest remnant (38.35 ±7.71) µm Mukwei Lagoon (17.98 ±5.25) µm	N/A	No identification of polymer types	fluorescent microscope	Not stated	Kpeshie Lagoon: 16.39±3.26 per 10 cm^3 of sediment, Mangrove forest remnant 25.94 ±3.13 per 10 cm^3 of sediment Mukwei Lagoon: 11.22 ±2.69 per 10 cm^3 of sediment	964
(Blankson et al., 2022)	2021	Fish Water Sediment	Weija Dam Densu Delta	water (1.208± 0.264)	N/A	No identification	microscope	Not stated	Weija Dam water: (1.583± 0.167) 60ml of water	Fish :57 Water: 14 Sediment: 31

				sediment (0.267± 0.051) Black-chinned Tilapia (0.605± 0.078) mm Bagrid Catfish (0.637± 0.061) mm		of polymer types			Densu Delta water (4.16 ± 0.342) 60ml of water Weija Dam sediment (3.75± 0.853) 40g of sediemt Densu Delta sediment (4.00± 0.408) 40g of sediment	
(Adu-Boahen et al., 2022)	Not stated	Fish Water sediment	River Akora, Central Region	Not stated	N/A	No identification of polymer types	Microscope	Not stated	Fish: 1.43 item / fish Water: 85 MPs loads per m^3	Water: 286 Fish: 30
(Faseyi et al., 2022)	2020-2021	Sediment	Pra Estuary Ankobra Estuary	Not stated	N/A	No identification of polymer types	Microscope	Fibres sheet fragment bead	Pra Estuary (728 /10g sediment) Ankobra Estuary 456 particles/10g sediment	1184
(Pappoe et al., 2022)	2020-2021	fish	Jamestown fish landing beach	0.1 – 1.0 mm	Polyethylene (PE), polyvinyl acetate (PVA) polyamide (PA)	Polymer were characterised	Microscope FT-IR	Fibres Pellets	*S. maderensis* (1.49 ± 1.48 items/fish) *P. prayensis* (1.26 ± 1.67 items/fish), *D. rhonchus* (0.96 ± 1.055 items/fish) *P. bellottii* (0.94 ± 1.18 items/ fish).	133

(Fadare et al., 2021)	2021	Table- salt	Nigeria Cameroon Ghana Malawi Zimbabwe South Africa Kenya Uganda	(3.3 to 4660) µm	polyvinyl acetate (PVA), polypropylene (PP) and polyethylene (PE)	Polymer were charaterised	Microscope FT-IR	fragments fibres, granules	(38.42 ± 24.62 particles/kg of salt).	1328
(Addo et al., 2022)	2021	Oyster	Volta Estuary Densu Estuary Nakwa Estuary Whin Estuary	(50 -1000) µm	polyethylene polypropylene polyamide polystyrene cellophane polyester	Polymer were charaterised	Microscope FT-IR	Film Fragment Fibre	Densu: (3.4 ± 1.0 items/individual) Volta: (2.8 ± 1.1 items/individual) Nakwa: (1.4 ± 1.3 items/individual) Whin: (1.6 ± 1.2 items/individual)	276
(Nuamah et al., 2022)	2021	Water	Coastal sea: Sekondi, Cape Coast Tema Denu	(2 – 5) mm	polypropylene (PP) polyethylene (PE) polystyrene (PS).	Polymer were charaterised	Microscope FT-IR	Fragment foam film pellet fibre	Cape Coast: 1.14 ± 0.63 particles m^{3} Sekondi: 1.33 ± 1.02 particles m^{-3} Tema: 2.79 ± 2.79 particles m^{-3} Denu: 2.36 ± 1.95 particles m^{-3}	2057

Source: cited papers in Table 2.