UNIVERSIDADE TÉCNICA DO ATLÂNTICO
INSTITUTO DE ENGENHARIA E CIÊNCIAS DO MAR

WEST AFRICAN SCIENCE SERVICE CENTRE ON CLIMATE CHANGE AND ADAPTED LAND USE

Master Thesis

ASSESSING THE STATE OF MICROPLASTICS POLLUTION LEVELS IN FISHES AND FISHING GROUNDS IN CABO VERDE USING MICROSCOPY AND A HYPERSPECTRAL IMAGING SYSTEM TECHNIQUE

WISE GOODLUCK DATSOMOR

Master Research Program on Climate Change and Marine Sciences

São Vicente
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Wise Goodluck Datsomor

Master's thesis presented to obtain the master's degree in Climate Change and Marine Sciences, by the Institute of Engineering and Marine Sciences, Atlantic Technical University in the framework of the West African Science Service Centre on Climate Change and Adapted Land Use

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Wise Goodluck Datsomor

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Dedication

With profound gratitude, I dedicate this work to God Almighty, the source of all wisdom and knowledge. I am eternally grateful for his endless love and favor, which saw me through all the days of my life until now.

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Resumo

Os microplásticos (MP) têm sido amplamente estudados pelos seus efeitos prejudiciais no ambiente. Este estudo investiga a ocorrência e distribuição de MP em espécies de peixes seleccionadas e em pesqueiros próximos à ilha de São Vicente, Cabo Verde, utilizando microscopia e imagem hiperespectral (HSI). Os resultados demonstraram uma presença predominante de partículas de MP, sendo as fibras a forma mais abundante e uma proporção significativa de MP de cor preta. Os padrões de distribuição de MP em forma e cor foram semelhantes em todas as estações de amostragem, não indicando nenhuma agregação específica. Enquanto os bancos de pesca da Mindelo 1, São Pedro e Calhau exibiram concentrações comparáveis de MP, Mindelo 2 apresentou níveis mais elevados. Aproximadamente 51% das amostras de peixes mostraram ingestão de MP, com *Caranx crysos* exibindo a maior abundância. Os peixes não demonstraram seletividade com base na forma ou cor, sugerindo a ingestão com base na disponibilidade. Não foram observados efeitos adversos significativos nas condições de crescimento dos peixes, apesar da ingestão de MP. A análise de polímeros revelou dois grupos proeminentes, Polietileno (PE) e Polipropileno (PP), indicando fontes potenciais de gestão inadequada de resíduos por parte dos indivíduos, bem como actividades de pesca. No entanto, as limitações do estudo incluem a incapacidade do sistema HSI para analisar amostras a preto e branco. É necessária mais investigação sem estas limitações para compreender de forma abrangente a poluição por MP na região de estudo.

Palavras-chave: Microplásticos (MP), Imagem hiperespectral (HSI), Microscopia, Polímeros

Abstract

Microplastics (MP) have been extensively studied for their detrimental environment effects. This study investigates the occurrence and distribution of MP in selected fish species and fishing grounds near São Vicente, Cabo Verde, utilizing microscopy and hyperspectral imaging (HSI). The results demonstrated a prevalent presence of MP particles, with fibers being the most abundant shape and a significant proportion of MP appearing black. MP distribution patterns in shape and color were similar across all stations, indicating no specific aggregation. While Mindelo 1, São Pedro, and Calhau fishing grounds exhibited comparable MP concentrations, Mindelo 2 had higher levels. Approximately 51% of fish samples showed MP ingestion, with *Caranx crysos* exhibiting the highest abundance. Fish did not demonstrate selectivity based on shape or color, suggesting ingestion based on availability. No significant adverse effects on fish growth conditions were observed despite MP ingestion. Polymer analysis revealed two prominent groups, Polyethylene (PE) and Polypropylene (PP), indicating potential sources from inadequate waste management by individuals as well as fishing activities. However, limitations of the study include the HSI system's inability to analyze black and white samples. Further research without such limitations is necessary to comprehensively understand MP pollution in the study region.

Keywords: Microplastics (MP), Hyperspectral Imaging (HSI), Microscopy, Polymer.

Abbreviations and acronyms

MP	Microplastics
HSI	Hyperspectral imaging
CLEAR	Civic Laboratory for Environmental Action Research
KOH	Potassium Hydroxide
MDS	Multidimensional Scaling
PE	Polyethylene
PP	Polypropylene

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1. Introduction

Microplastics (MP) are ubiquitous and have become a global concern, particularly in marine environments, due to their widespread occurrence and persistent nature (Thompson et al., 2004). The potential adverse effects of MP on both ecosystems and human health have raised significant alarms (Barboza et al., 2018). MP are defined as artificial polymeric particles that typically exhibit moldability under heat and pressure (Rodriguez, 2018). They vary in size, ranging from 1 µm to 5 mm in length (Barnes et al., 2009; Hidalgo-Ruz et al., 2012; Thompson et al., 2004) and originate from diverse sources. Primary MP are intentionally produced in the micrometer range for specific purposes or are released from plastic products during use, e.g., from clothing or from car tyres. Secondary MP results from the fragmentation and degradation of larger plastic items (Hurt et al., 2020).

Numerous scientific investigations have extensively explored the occurrence and distribution patterns of MP in diverse fish species inhabiting a wide range of aquatic environments, encompassing both pelagic and demersal habitats (Barboza et al., 2020; Koongolla et al., 2022; Mahu et al., 2023; McNeish et al., 2018; Murano et al., 2020; Smith et al., 2018; Watts et al., 2016). These studies have revealed the presence of various types of MP, including microbeads, fibers, films, nurdles/pellets, fragments, and threads, within multiple anatomical regions of fish, such as the digestive systems, gills, tissues, and liver (Adika et al., 2020; Mahu et al., 2023; McGoran et al., 2017). These findings provide compelling evidence for the pervasive nature of MP contamination in fish populations.

The wide prevalence of MP in fish can be attributed to a combination of factors, primarily influenced by the biological activities of these organisms, including their feeding habits and specific habitats (McNeish et al., 2018). It has been observed that adult fish generally exhibit higher concentrations of MP in their tissues compared to their juvenile counterparts, indicating a potential age-dependent ingestion pattern (Lusher et al., 2017; McNeish et al., 2018). This finding suggests that the accumulation of MP may increase over time as fish grow and interact with their environment.

Ingestion of MP is the most likely interaction between life forms and MP and it occurs among a wide range of species, from low trophic level organisms to predators, including invertebrates, fishes, mammals, turtles and seabirds (Cole et al., 2013; Desforges et al., 2015; Lusher, 2015; Nadal et al., 2016). Organisms often mistake these particles for food and indiscriminately feed on them due to the feeding strategies employed by some of these organisms (Besseling et al., 2015; Hall et al., 2015). This can lead to the internal blockage of the gills and the digestive tract of fish (Białowąs et al., 2022). The uptake of food by these organisms would then decline since they feel satisfied anytime they mistake plastics for food, resulting in poor energy supply, low growth and decreased fecundity (Besseling et al., 2013; Sussarellu et al., 2016). Toxicity tests have highlighted the adverse physical (Wright et al., 2013) and toxicological effects that MP exposure can have on marine biota (Cole et al., 2015; Ogonowski et al., 2016; Pedà et al., 2016; Watts et al., 2016). Experiments using marine worms and zooplankton have demonstrated that MP ingestion can result in reduced feeding, decreased growth rates, increased mortality, decreased hatching success and reduced fecundity (Cole et al., 2015; Wright et al., 2013). To a greater extent, there is evidence of MP causing neurotoxic and oxidative damage in wild fish (Barboza et al., 2020). Furthermore, MP can adsorb and transport chemical pollutants, which can bioaccumulate and biomagnify in the food web (Carbery et al., 2018).

The potential risks associated with MP extend beyond marine ecosystems and can affect human health (Smith et al., 2018). MP can enter the human body through various pathways, including ingesting contaminated seafood, drinking water, and inhalation of airborne particles (Smith et al., 2018). The long-term effects of chronic exposure to MP on human health are not yet fully understood and require further investigation (Smith et al., 2018).

The archipelago nation of Cabo Verde, located in the eastern Atlantic Ocean, represents one of the biologically diverse regions that serves as one of the world’s most important life support systems. Its coastal waters serve as substantial fishing grounds and support the archipelago with seafood for both domestic consumption and export. With a potential fish catch varying between 36,000 tons and 44,000 tons, a yearly catch of 10,000 tons is achieved, providing about 11.2 kg/per capita/year (*Cabo Verde | Spcsrp | Sub Regional Fisheries Commission*, n.d.; FAO, 2020). The possible introduction of MP into this oceanic system by major current systems such as the North Atlantic Subtropical Gyre (Cardoso & Caldeira, 2021), as well as local sources including inadequate waste management, coastal urban areas, and fishing activities, can have detrimental consequences for both the environment and the local fishing industry, significantly impacting the marketability and consumption of fish products.

Although, there is a growing global concern about MP pollution in oceanic systems, so far only very limited research has been conducted on the occurrence and distribution of MP in seafood and the fishing grounds of Cabo Verde (Fernandes, 2019; Ferreira et al., 2021). However, knowing the levels and the spatial and temporal patterns of MP contamination in this region is crucial for assessing potential risks to both the environment and human populations that rely on seafood consumption (De-la-Torre, 2020).

This master's thesis serves as a baseline study that aims to address this research gap by employing microscopy and a hyperspectral imaging (HSI) system technique to assess the levels and distribution of MP in the stomach contents of five of the economically important fishes and in water samples from different fishing grounds along the coastline of São Vicente, Cabo Verde. The findings from this study will contribute to the understanding of MP contamination in this region and provide valuable insights and recommendations for further research.

1.1 Aims and Objectives

This research will ascertain the distribution and occurrence of MP in five selected fish species and 4 fishing grounds in São Vicente Island of Cabo Verde employing microscopy for MP detection and a HSI system technique for identifying the chemical nature of the detected particles. To achieve this, the following specifics objectives were set:

- To identify and quantify the number, colour, and shapes of MP in the guts of the fishes and sampled waters;
- To canvass the correlation between the fishes' condition factor (length and weight parameters) and the total number of MP in fish guts and seawater samples, respectively;
- To determine the polymer composition of MP (down to a minimum size of 300 µm in length) from fish and collected seawater samples.

2. Literature review

The ocean is significantly impacted by various forms of pollution, leading to the disruption of the food web, habitat destruction, the eventual death of organisms and potentially the extinction of species (Singh & Singh, 2017). Plastic pollution is one of such form of pollution that has gained global attention due to its prevalent occurrence in marine ecosystems and its profound effects on fish, mammals, seabirds, and invertebrates in the marine environment (Auta et al., 2017). It is considered a global issue, with its magnitude of impact being comparable to climate change (UNEP, 2018).

Plastic pollution, specifically microplastics (MP), is prevalent throughout the water column, on the seafloor, and across all habitats in both open and enclosed oceans (Auta et al., 2017). The study of MP is of great concern in marine and environmental science (Alfaro-Núñez et al., 2021) and has seen significant growth over the past decade (Barboza & Gimenez, 2015). However, most of these studies have focused on European waters, resulting in a disparity in the geographic coverage of research (Klingelhöfer et al., 2020). Consequently, there is a lack of data on MP studies in Africa.

This section provides a comprehensive overview of existing knowledge, research, and findings related to the occurrence and distribution of MP in seafood and fishing grounds.

2.1 Microplastics – Definition, sources, occurrence and distribution

Plastics play a pivotal role in various aspects of human life, encompassing domestic, industrial, agricultural, medical, transportation, packaging, and many other fields (Bhat et al., 2023; Mitrano & Wagner, 2022; Wei & Zimmermann, 2017). Their widespread utilization stems from their exceptional versatility, high durability, cost-effectiveness, hygienic packaging properties, resistance to water and certain chemicals, superior thermal and electrical insulation, and lightweight nature compared to alternative materials (Hatow, 2006; Mitrano & Wagner, 2022; Thompson et al., 2009). These plastic materials are produced via polymerization, a process involving the application of heat and pressure. Bahraini (2018), conducted a study classifying plastics into different types based on their polymer structures, including polyethylene terephthalate (PET/PETE or polyester), high-density polyethylene (HDPE), polyvinyl chloride (PVC), low-density polyethylene (LDPE), polypropylene (PP), polystyrene (PS), and a miscellaneous category encompassing other plastic types formed through polymer combinations, exemplified by Polycarbonate. However, despite their immense importance, mismanaged plastics have significant environmental impacts, largely due to pollution resulting from population growth, modernization through industrialization and the increasing demand for plastics in any form (Nathanson, 2019). According to Geyer et al. (2017), the total global mass of plastics produced since the 1950s is estimated to be approximately 8 billion tons. Alarmingly, around 60% of these plastics has been discarded into the environment, either placed in landfills or into the wild, where it gradually breaks down into smaller fragments.

MP, defined as minute plastic fragments typically measuring less than 5 mm in length, pose grave threats to the marine ecosystem (National Oceanic and Atmospheric Administration, 2018). These MP can be classified into two main types: primary MP, which are intentionally produced in laboratories (e.g., microbeads and micro pellets), and secondary MP, which forms from the degradation of larger plastics under the influence of UV radiation, wave actions, and other factors. They are ubiquitous and have been documented in all compartments of the planet including the atmosphere, ocean, rivers, and in living organisms (Auta et al., 2017; Lim, 2021).

Studies have revealed the presence of MP particles in various human samples, including the placenta and unborn babies (Ragusa et al., 2021), as well as in commonly consumed food items such as honey, salts, milks, and drinking water (Gambino et al., 2022; Kutralam-Muniasamy et al., 2020; Peixoto et al., 2019). Furthermore, MP have been detected in human blood and stool samples (Leslie et al., 2022; Schwabl et al., 2019).

The ocean is a significant reservoir for MP, acting as the largest sink for these particles (Rochman, 2018). Astoundingly, it is estimated that the surface of the world's oceans contains approximately 82-358 trillion plastic particles, weighing around 1.1-4.9 million tonnes (Eriksen et al., 2023). This highlights the extensive presence of MP and their accumulation in marine ecosystems.

Rivers play a crucial role in transporting plastics from land-based sources, including landfill refuse, sludge, and food waste, into the marine environment (Golwala et al., 2021; Y. Li et al., 2020). Moreover, estuaries have been identified as MP hotspots, as these transitional environments are highly susceptible to plastic contamination (Bergmann et al., 2015). Within estuaries, hydrodynamic forces such as currents, tides, winds, and waves act upon MP, facilitating their distribution and dispersal into the marine environment (H. Zhang, 2017). Due to their proximity to MP sources, freshwater and transitional environments, including estuaries, serve as important pathways for transporting MP into the oceans (Dris et al., 2020). Also, one of the often-overlooked and probably the most incredible routes of MP transport into the ocean is the atmosphere, which is estimated to transport about 0.013 to 25 million metric tons of MP annually into the oceans (Allen et al., 2022; Evangeliou et al., 2020). Once these particles reach the ocean, they interact with marine ecosystems from the surface to the seafloor, with partly significant and adverse impacts (Jeong et al., 2017; Paul & Isaac, 2023).

2.2 The impacts of microplastics on marine ecosystems

The presence of MP throughout various parts of the marine environment has significant implications for both the short-term and long-term health of marine ecosystems. This is due to their persistent nature and widespread distribution (Barnes et al., 2009; Lusher, 2015). As these synthetic particles integrate into the environment, they interact with both living and non-living elements (Galloway et al., 2017). One critical interaction involves the ingestion of MP by organisms across different trophic levels, from zooplankton to higher-level consumers such as fishes, seabirds and marine mammals, as they are often mistaken for food (Cole et al., 2013; Desforges et al., 2015; Lusher, 2015; Nadal et al., 2016).

Research has demonstrated the harmful consequences of MP ingestion on organisms (Prinz & Korez, 2019). Ingestion can result in physical and internal injuries such as intestinal damage. Furthermore, it can disrupt feeding behaviors, hinder digestion, impair reproduction and growth, and ultimately affect population dynamics and biodiversity (Besseling et al., 2013; Cole et al., 2013; Ogonowski et al., 2016; Sussarellu et al., 2016; Wright et al., 2013). These impacts occur as MP interfere with normal physiological processes and introduce foreign substances into the organisms' bodies (Agathokleous et al., 2021). Also, MP have the potential to bioaccumulate and biomagnify within the food web (Besseling et al., 2013; Rajesh, 2020). Organisms that ingest MP can experience the accumulation of toxic substances adsorbed onto the plastic surfaces (Cole et al., 2015; Teuten et al., 2009). As predators consume contaminated prey, the accumulated MP and associated pollutants can be transferred up the food chain, reaching higher trophic levels and posing risks to both wildlife and human health (Nelms et al., 2019).

Aside from ingestion by single species, MP can potentially disrupt critical ecosystem functions and processes. According to Shen et al. (2020), MP can interfere with ocean carbon sequestration by altering the marine primary production, since their presence in the ocean can affect phytoplankton photosynthesis and growth by reducing light transmission. Also, these MP particles can serve as a substrate for phytoplankton aggregation thereby reducing light availability and impacting productivity (Amaral-Zettler et al., 2020; Shen et al., 2020; Sjollema et al., 2016). Additionally, MP can sink to the ocean floor due to their density and biofilm coating, affecting the circulation of organic matter and nutrients in deep waters which could potentially impact the carbon stock of the ocean (Shen et al., 2020). The ocean biological and microbial carbon pump, which are the main ways of ocean carbon sequestrations, are also disturbed by the presence of MP as they interfere with the fecal pellets (which transport carbon to deeper waters) through contamination, thereby reducing their descent flow and the proportion of sequestrated carbon (Wieczorek et al., 2019).

Furthermore, the accumulation of MP in habitats alters sediment characteristics, disrupts benthic communities, and interferes with nutrient cycling (Vegter et al., 2014). Fragile ecosystems like coral reefs and seagrass beds are particularly vulnerable, compromising their structure and function (Houseago et al., 2023; Vegter et al., 2014). MP also impacts filtration rates of bivalves and infaunal invertebrate communities in sediments (Salerno et al., 2021). All these impacts of MP on ecosystems can be further exacerbated by interactions with other environmental stressors (Lehmann et al., 2019). When MP interact with chemical contaminants, their combined effects can lead to additive or synergistic toxicity, amplifying the ecological risks they pose (Cao et al., 2021; Varg & Svanbäck, 2023).

While interacting with environments contaminated with MP, humans are not exempt from potential health implications (Yang et al., 2022). However, the available evidence is limited, and further research is needed to draw conclusive findings on the specific health effects induced by MP exposure in humans (Koelmans et al., 2022).

2.3 Challenges and knowledge gap in microplastics studies

Research on MP has escalated over the last decades in the quest to comprehend the full context of this menace. However, understanding MP distribution and impacts remains incomplete due to several challenges (Golwala et al., 2021; Mao et al., 2022; Xue et al., 2020).

One of such challenges has to do with the disparities in research coverage. While certain regions have received substantial attention, others, such as Africa, Central Asia, Southeast Asia, and South America, have been relatively understudied (Adeogun et al., 2020; Dong et al., 2021; Yücel et al., 2023).

Furthermore, the lack of standardized methods in MP studies also poses a significant challenge for accurately assessing and comparing data across different studies and environments (Baldwin et al., 2020; Biyik & Baycan, 2021; Hermsen et al., 2018; Hidalgo-Ruz et al., 2012; Renner et al., 2021). Standardized protocols for sampling, pretreatment, identification, and quantification are crucial for reliable and reproducible studies (Hidalgo-Ruz et al., 2012; Renner et al., 2021). Implementing such protocols is vital for monitoring MP pollution, understanding its impacts, and assessing environmental and human health risks (Ivleva, 2021).

In addition, the focus on the marine environment has often marginalized the context of terrestrial sources such as rivers, lakes, and estuaries (Galloway et al., 2017; Golwala et al., 2021). This has resulted in inadequate information regarding the quantification of sources and pathways of MP, making it difficult to assess the detailed ecological and health impacts of these particles (Cole et al., 2015; Koelmans et al., 2019).

2.4 The plastic problem in Cabo Verde

Cabo Verde is one of the 5 Macaronesia archipelagos situated about 570 km from the Western coast of Africa and in the Northeastern Atlantic Ocean (central) with a total coastline of about 965 km. Plastic pollution is one of the major concerns although only a few studies are publicly available (Catarino et al., 2023; Fernandes, 2019; Ferreira et al., 2021; Osemwegie et al., 2021; Sousa-Guedes et al., n.d.; Weidlich & Lenz, 2022). Plastics are almost inevitable in people's daily lifestyles with almost every household using plastics. The lack of adequate and improper waste management systems, which was reported by the Ministry of Environment, Agriculture and Fisheries in the year 2004 could be one of the sources of plastic pollution. Ferreira et al. (2021), noted that the behavior of individuals in handling plastics is a major cause of plastic pollution in this area. Additionally, Cardoso & Caldeira (2021) demonstrated, by using modeled datasets for ocean currents, winds, and waves with a Lagrangian tool, that sources of the plastic pollution in the Cabo Verdean marine environment originate from the Northwestern African coast (Morocco, Libya, Tunisia, Algeria). Other studies, especially from Fernandes (2019), indicate large amounts of plastics ranging in different shapes and fragments dominating debris collected along the Praia dos Achados beach situated on Santa Luzia island - an isolated island. MP ranging from fibers, fragments, and pellets were also identified in some fishes (gut, muscle, and gills) sampled (Fernandes, 2019).

Fujisaki & Lamont (2016), highlighted some effects of plastics pollution along shores and beaches on sea turtles, where they are entangled to death and have their nesting behavior altered. The success of hatchlings reaching the ocean is marginalized due to their interaction with plastic debris (Triessnig et al., 2012).

In light of this pollution menace, numerous actions and events including public awareness on the impacts of plastics, and beach clean-ups, among others have been implemented by

Organizations like Biosfera I, Sal a Pente Fino, Calao Luxembourg A.S.B.L. However, these activities are just in the developing states. To have a better understanding of the situation and better assess the health implications of MP within this region, research is needed, hence the rationale behind this thesis.

3. Material and methods

3.1 Study area

Cabo Verde, situated in the central Atlantic Ocean (16.5388° N, 23.0418° W), is an archipelago country comprising ten (10) volcanic islands with a total land area of approximately 4,033 square kilometers. It has a coastline of 965 kilometers, which exhibits diverse coastal features, including sandy and rocky beaches, dunes, and low-lying rocky coasts (Rosa et al., 2023; Gomes et al., 2019) (Figure 1). The coastline is influenced by the Canary Current, which flows along the eastern border of the Azores anticyclone and connects with northern trade winds (Freitas et al., 2019; Hernández-Guerra et al., 2002; López et al., 2019). These oceanographic patterns including the Gulf Stream and North Atlantic subtropical gyre play a crucial role in facilitating the interchange of species, the transfer of pollutants (including MP), and fostering connectivity between Cabo Verde and its neighboring regions (Cardoso & Caldeira, 2021). The coastal waters of Cabo Verde are renowned for their rich marine biodiversity, harboring endemic species of coastal fishes, gastropods, brachyurans, and polychaetes (Freitas et al., 2019). Moreover, the coastline experiences the impact of Saharan dust, which is transported over long distances and influences regional atmospheric conditions, as highlighted by Rittmeister et al. (2017). These dust particles can transport MP particles from afar into the environment, polluting the waters of Cabo Verde (Brahney et al., 2021).

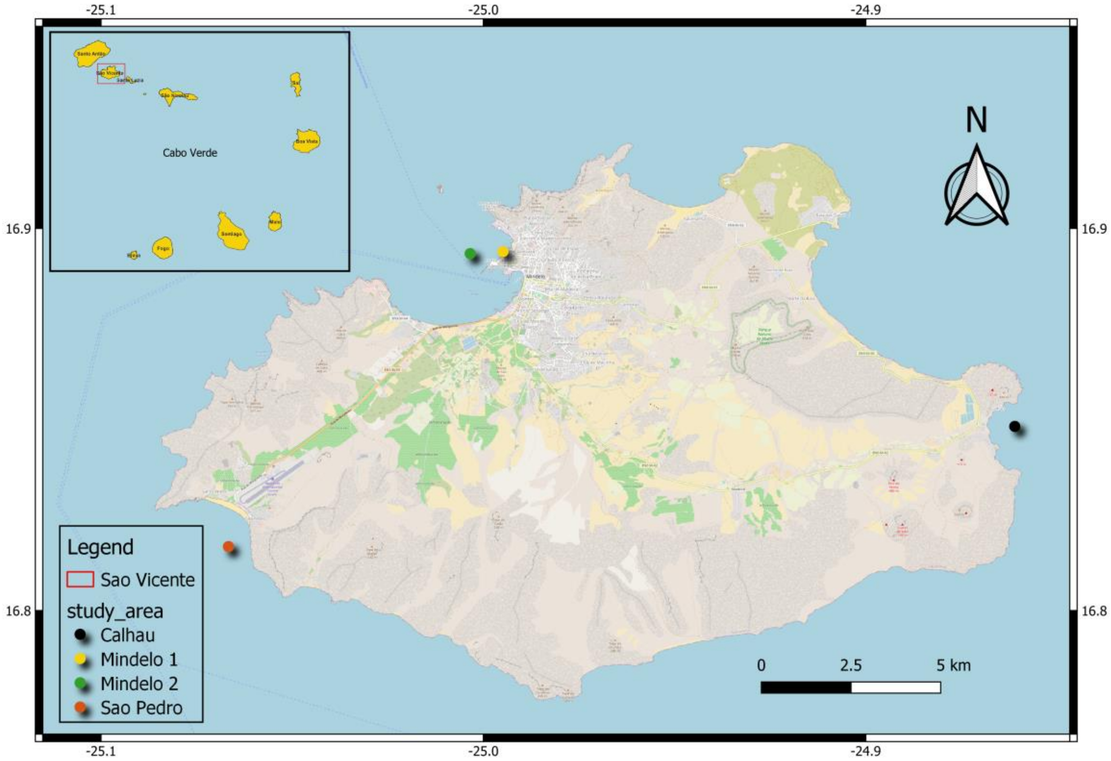

Figure 1: Map of study area highlighting stations for the microplastic net tows (by Wise Goodluck Datsomor).

Among the ten (10) islands of the Cabo Verde Archipelago, São Vicente, located at 16˚51"N 24˚58" W, is part of the Barlavento Islands and stands out for its cultural significance and tourism potential, because of its picturesque landscapes, including rocky terrain, rolling hills, and beautiful beaches (Zhuang et al., 2019). Due to its prominent geographical location and distinctive attributes, it draws the attention of researchers from around the globe who are intrigued by its cultural dynamics, coastal ecosystems, and socioeconomic aspects (Zhuang et al., 2019). Mindelo, the most developed and inhabited town on the island, houses approximately 92% of the total population, while other towns such as Baía das Gatas, Lameirão, Monte Verde, Norte de Baía, Ribeira de Vinha, Ribeira de Calhau, Ribeira de Julião, Salamansa, and São Pedro contribute to the island's demographics. Besides tourism, the bustling Porto Grande, the main port of Cabo Verde plays a significant role in generating income (Carolyn, 2022)

The climate over São Vicente Island limits agricultural activities due to low precipitation, resulting in subsistence-level farming. However, fishing serves as a local income-generating source, with the Mercado de Peixe as the buzzing and busy fish market filled with fish mongers almost daily (Fortes et al., 2019)

The issue of marine pollution is a concern and cannot be hidden although not much research is publicly available (Ferreira et al., 2021). Considering the threats and impacts of plastic pollution, it is prudent for this research to be undertaken to bewray the extent of MP pollution in some fishes and seawater samples here in São Vicente Island.

3.2 Field sampling

3.2.1 Fish samples

A total of one hundred (100) fish individuals, representing five different species, were obtained from the fish market (Mercado de peixe) in Mindelo on the early mornings of 16th and 17th March 2023, shortly after being landed by local fishers. The selected fish species included 20 individuals each of *Caranx crysos, Cephalopholis taeniops, Decapterus macarellus, Lithognathus mormyrus,* and *Selar crumenophthalmus*. These species were specifically chosen due to their recognized importance as commonly consumed fishes in Cabo Verde (R. Freitas, Personal Communication, March 27, 2022).

Upon acquisition, the fish samples were immediately placed in an ice-filled cooler to preserve their freshness and transported to the laboratory at the Instituto do Mar (IMar). In the laboratory, the fish samples underwent appropriate preparations by being thoroughly washed under running Milli-Q water to facilitate subsequent analysis and investigations. This process ensured that the samples were kept in good condition and to prevent MP contamination before further scientific examination.

3.2.2 Water samples

The sampling of MP in this study involved the selection of four fishing grounds, namely Mindelo 1, Mindelo 2, Calhau, and São Pedro, based on consultations with local fishers who indicated these areas as locations where the selected fish species are commonly caught. Sampling was conducted using a local artisanal fishing vessel that towed a MP trawl net that was equipped with a flowmeter. The trawl net had an opening area of 0.28 square meters and a mesh size of 300 µm, which was securely fixed behind the vessel at a distance of approximately 2 meters.

Sampling activities took place along five defined transects within each study site. At each transect, MP samples collected from the net tow were transferred into two well labeled glass bottles (500 ml). The precise GPS coordinates and time were meticulously recorded at the beginning and end of each tow to ensure accurate spatial and temporal information. In addition, the flowmeter readings were noted to calculate the total volume of seawater sampled during each tow. The duration of each tow averaged approximately 10.20 minutes, with an average volume of 110.8 m^3 of seawater towed at a speed of 2 knots (3.70 km/h).

To maintain sample integrity, the collected MP samples were immediately stored in an ice cooler during fieldwork and subsequently transported to the wet laboratory of the Ocean Science Centre Mindelo (OSCM) for laboratory analysis. The laboratory analysis aimed to quantify and characterize MP particles present in the collected samples.

The sampling campaign was conducted over a four-day period (13th, 14th, 15th and 18th March 2023), with each fishing ground being sampled within specific time frames. Sampling activities were conducted from late mornings to early afternoons, specifically between 11:30 and 15:00 GMT-1. Each fishing ground was sampled for approximately 2 to 3 hours daily, comprehensively assessing MP presence and distribution patterns in the designated areas.

3.3 Laboratory analysis

So far, several methodologies have been tested for the identification and characterization of MP (Lusher et al., 2016). They span from the use of the hot needle method for aiding visual identification during microscopy to more sophisticated methods such as Raman Spectrometry, Attenuated Total Reflectance (ATR), and Fourier Transformed Infrared Spectrometry (FT-IR) (Lusher et al., 2016) which are technique that uses infrared light to examine the unique spectral properties of MP particles, allowing for the identification and characterization of their polymer composition by measuring absorption and transmission.

In this research, a combination of morphometric measurements (length and weight) was taken to assess the condition of the fish. Additionally, the fish samples were dissected, and their guts were digested and filtered. The filtrates were visually inspected, using the hot needle approach as employed by Lusher et al. (2017). Furthermore, the CLEAR spotters guide, a document that provides guidance on identifying and classifying MP particles into various forms (Liboiron, 2017), was used to identify and classify the MP particles. Additionally, to further analyze the polymer makeup of the MP particles down to a minimum size of 300 µm in length, a hyperspectral imaging (HSI) system, as described by Beck et al., (2023), was employed. This advanced imaging technique allowed for the detailed examination and characterization of the MP samples in terms of their polymer composition. The following subsections provide a detailed elaboration of the various steps, organized into the following groups:

- Morphometric measurement, and dissection.
- Digestion and filtration
- Observation and identification of MP
- Polymer identification.

3.3.1 Morphometric measurement and dissection

At the IMar fish laboratory, fish individuals were thoroughly washed with Milli-Q water to prevent potential secondary contamination with plastics. Each individual fish was measured to obtain the total body length (cm) and standard length (cm) using a fish measuring board. Additionally, each fish's body weight (g) was recorded using an electronic weighing balance (top pan balance).

Utilizing a dissecting kit, a precise dissection procedure was carried out on each fish, starting from the anal opening and extending to the anterior region. The entire gastrointestinal tract (gut) was carefully extracted. The weight of the gut was measured using the top pan balance and kept in a labeled and tightly closed glass test tube and prepared for the subsequent digestion process. These steps were executed consistently for all fish individuals within a highly sanitary environment to ensure accuracy and reliability of the results.

3.3.2 Digestion and filtration

The digestion process aims to thoroughly break down all organic constituents present within the fish guts, resulting in the retention of solely inorganic substances, which potentially encompass MP. The use of various chemicals, like nitric acids (HNO_3), hydrochloric acids (HCl), hydrogen peroxides (H_2O_2), sodium hydroxide (NaOH), and others, in the digestion of various parts of the fish including the stomach, tissues, oesophagus, intestines etc. have had varying results concerning digestion efficacy. Nitric acid (HNO_3) exhibited notable effectiveness in digesting biological tissues (Lusher et al., 2017). However, it has also been observed that the digestion process can sometimes leave behind oily residues and residual tissues, which could obscure MP and impede visual identifications, leading to errors (Claessens et al., 2013; Cole & Galloway, 2015; Santana et al., 2016). Other studies have employed further methodologies, including enzymatic digestion, various concentrations of sodium hypochlorite, oxidizing agents, and alkaline substances, but Lusher et al. (2017) as well as Dehaut et al. (2016) found that using 10% potassium hydroxide (KOH) and incubating at 60 ºC overnight yielded highly successful results in the digestion of fish guts.

Consequently, a solution of one (1) liter of 10% KOH was prepared by dissolving 100 g of KOH salt pellets in 1 L of Milli-Q water. Subsequently, 20 ml to 50 ml of the prepared 10% KOH solution was added to the fish guts in the labeled glass test tubes. These tubes, containing the guts and the added 10% KOH solution, were then incubated in an oven at 60 ºC for 12 h to ensure complete digestion.

Similarly, in the case of the seawater samples, highly turbid samples, i.e. those that presumably contained a large amount of organic material, were subjected to digestion by adding 200 ml of 10% KOH to 500 ml of seawater and subsequently incubating them overnight at 60 ºC (Kye et al., 2023; Mbachu et al., 2021). Unfortunately, some water samples did not exhibit complete digestion, but displayed the accumulation of white precipitates at the bottom of the bottles. These samples were meticulously decanted, and the precipitates were dissolved using a 20 ml solution of 10% hydrochloric acid (HCl), allowing for an overnight reaction. This approach led to the disappearance of all precipitates, indicating the complete digestion of all organic components.

The digested fish guts and water samples were individually filtered through a moistened 1.2 µm and 0.45 µm Whatman GF/C microfiber filter paper respectively which were placed in the filtration setup. Subsequently, the filter papers were kept individually in a well labelled and covered glass petri dishes. They were then dried in an oven at 40 ºC overnight and prepared for visual identification with a microscope.

3.3.3 Observation and identification of microplastics

The dried filter papers with filtrates from both sample types were subjected to examination under a Kern Optics OZP 558 Stereo zoom microscope equipped with an integrated ODC 832 digital camera. Accompanied by VIS 2.0 Pro software, an application serving as an interface between the microscope and the desktop monitor, MP particles were identified and subsequently classified following the CLEAR spotter’s guide (Liboiron, 2017) (Figure 2) into diverse plastic types, encompassing microbeads, fibers, fragments, threads, films, foams, pellets, as well as distinct colors such as white, green, red, black, and blue. The software's operational capacity facilitated measurements and the capture of pertinent images for further scrutiny. This methodology was consistently applied to all samples.

Figure 2: The CLEAR Spotter's guide that provide details on microplastics identification adopted from Liboiron., (2017).

3.3.4 Polymer identification

For the characterization of polymer types, MP particles greater than 300 µm in length were picked with clean forceps and arranged on a black microscopy slide which was then placed under a Specimen FX17 camera (Near InfraRed HSI System). The particles were then illuminated from overhead by two halogen lights positioned at approximately 45° from the front and back of the camera target field, respectively. Hyperspectral images were captured for further processing using the Lumo software suite (Beck et al., 2023).

To identify the particles on the hyperspectral images, they were processed using a custom software written in Python. It comprised a combination of the Sobel edge-finding algorithm and the watershed segmentation algorithm from scikit-image, an open-source collection of algorithm written in python language that helps in image processing (Walt et al., 2014). The Sobel filter was employed to detect pixel intensity gradients and locate the edges of the particles. The identified edges were used as seeds for the watershed segmentation algorithm, which then filled the pixels associated with each particle (Beck et al., 2023)

Before segmentation, the hyperspectral images underwent preprocessing by calculating the mean spectrum for each pixel. This step ensured proper formatting of the hyperspectral data. Singular value decomposition was then applied to determine the major and minor axis lengths of the particles, providing information about their shape and size (Beck et al., 2023).

The classification of particle polymer types was achieved using the scikit-learn random forest algorithm. This algorithm was trained on the one-hot encoded data, a process that represents categorical data as binary vectors. This encoding process converts each category into a binary feature, allowing the algorithm to accurately predict and classify based on the categorical information from the reference library. By considering the presence of peaks and troughs in the spectra as classification features, the algorithm learns patterns and relationships between the categories, enhancing its ability to make precise classifications. This approach enabled the accurate identification of particle polymer types.

3.4 Quality assurance

To ensure a good quality of the laboratory analysis, strict measures were taken to prevent contamination. This involved wearing a cotton laboratory coat, hairnet, and hand gloves throughout the analysis. Additionally, the use of plastic materials was minimized whenever possible. The work environment was maintained clean to minimize the risk of contamination. Prior to the experiment, all apparatus used were pre-cleaned using Milli-Q water. Metallic materials, such as dissecting kits, were wiped with ethanol to further reduce the chances of contamination. Reusable items, including test tubes, petri dishes, and forceps, were thoroughly washed and cleaned with tissue papers to prevent cross-contamination.

To safeguard against external contamination, the filtration process for all samples was carried out in a fume chamber. Moreover, strict adherence to standardized digestion and drying time durations was maintained consistently for all samples.

The methodology was validated by conducting sample experiments using a 10% KOH solution as a system blank before the main experiment. The inspection process employed a certified spotter guide from the CLEAR to ensure accurate identification and characterization of MP. Throughout the analysis, five blank filters were strategically placed in the laboratory to account for any potential contamination. Notably, no MP particles were detected during the analysis, affirming the absence of contamination.

For polymer classification, a set of virgin plastic polymer beads, namely polystyrene (PS), high-density polyethylene (HD-PE), low-density polyethylene (LD-PE), Polymethyl methacrylate (PMMA), polyamide 66 (PA66), polycarbonate (PC), polyethylene terephthalate (PET), and polypropylene (PP), were analyzed as standards. The results demonstrated accurate predictions, confirming the system's precision and ability to provide correct outcomes.

3.5 Statistical analysis

The obtained results are presented as mean ± standard error. To assess the significance of differences in fish morphometrics (length and weight) among different fish species, the number of MP ingested by fish species, and the number of MP sampled across various stations, a one-way analysis of variance (ANOVA) and Turkey Pairwise test were conducted. Prior to running the ANOVA test, the data underwent a normality test to ensure it followed a normal distribution. Using R and Python programming languages, various graphical representations were generated, including Multidimensional scaling (MDS) graphs, bar plots, box plots, strip plots, and combinations of strip and box plots.

The condition factor, which reflects the physical and biological circumstances and fluctuations through the interaction of feeding conditions and physiological factors (Ighwela et al., 2011), was calculated using the Fulton’s Condition Factor formula,

$$K=\frac{W}{L^3} * 100 \qquad \text{(Eq 1)}$$

where, K = Fulton’s Condition Factor, W= Total Weight (g) of fish and L= Total Length (cm) of fish. The condition factor (K) provides a quantitative assessment of the growth conditions within a population and can potentially indicate the relationship with MP ingestion by fish (Ighwela et al., 2011).

To assess the correlation between MP in fish and water samples, the moving average method was employed to align the length of the fish data with that of the water samples.

4. Results

4.1 Morphometry and condition factor of fish

The fish species studied showed wide ranges of length and weight. The calculation of the condition factor revealed that the individuals from almost all fish species were in good physiological conditions, except for *Decapterus macarellus*, which recorded condition factor values of less than 1 which meant they were not growing as they should. This observation suggests that a substantial portion of the sampled fish populations exhibit good health and proper nourishment (Table 1).

Fish Species	Range Length (cm)	Mean Length (cm)	Range Weight (g)	Mean Weight (g)	Mean Condition Factor K
Caranx crysos	25.0 to 29.5	27.48 ± 1.25	193.11 to 275.80	246.83 ± 25.91	1.19 ± 0.09
Cephalopholis taeniops	23.0 to 34.5	27.18 ± 2.97	166.22 to 500.30	276.92 ± 92.69	1.34 ± 0.12
Decapterus macarellus	29.0 to 37.5	32.90 ± 2.39	235.28 to 599.28	359.79 ± 95.60	0.98 ± 0.06
Lithognathus mormyrus	24.0 to 37.0	31.38 ± 4.38	177.47 to 584.73	397.10 ± 147.18	1.23 ± 0.07
Selar crumenopthalmus	23.5 to 26.5	24.95 ± 0.74	137.11 to 197.84	165.33 ± 15.52	1.06 ± 0.04

Table 1: Individuals of the different fish species differed significantly in length and weight.

4.2 Occurrence of microplastics particles

A total of 341 microplastics (MP) particles, encompassing microbeads, fibers, fragments, threads, films, foams, and pellets of varying colors including brown, blue, black, green, red, and white/transparent were detected in this study (Figure 3). These particles were identified in both fish gut and net tow samples. Among these, 51% of the fish gut samples contained MP (119 particles), indicating that for every 2 gut samples analyzed, one contained at least one MP particle. *Caranx crysos* individuals contained the highest MP (39 particles) while *Selar crumenophthalmus* contained the lowest with 16 particles (Figure 4). Fibers emerged as the most abundant form of MP across the fish gut samples (Figure 7).

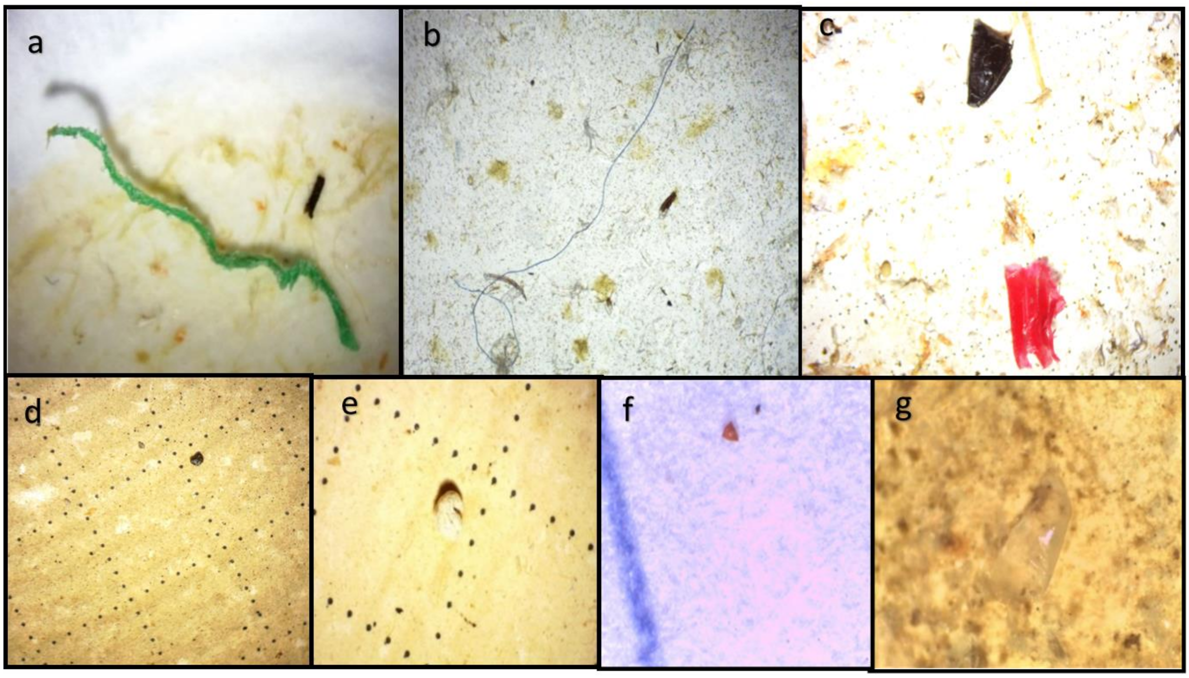

Figure 3: Various types of microplastics that were observed in either fish gut or water samples: a) thread, b) fiber, c) fragments, d) microbead, e) foam, f) pellet and g) film.

From a total volume of 2216 m^3 of seawater sampled across all stations (equivalent to 554 m^3 at each station), 222 MP particles were identified. Among the stations, Mindelo recorded the highest count of MP particles with 77 particles, while São Pedro had the lowest count (45) (Figure 5). Similar to the MP composition in the fish guts, fibers were the predominant form observed in the water samples (Figure13).

These findings highlight the widespread presence of MP in both fish and water samples, with fibers being the most prevalent form.

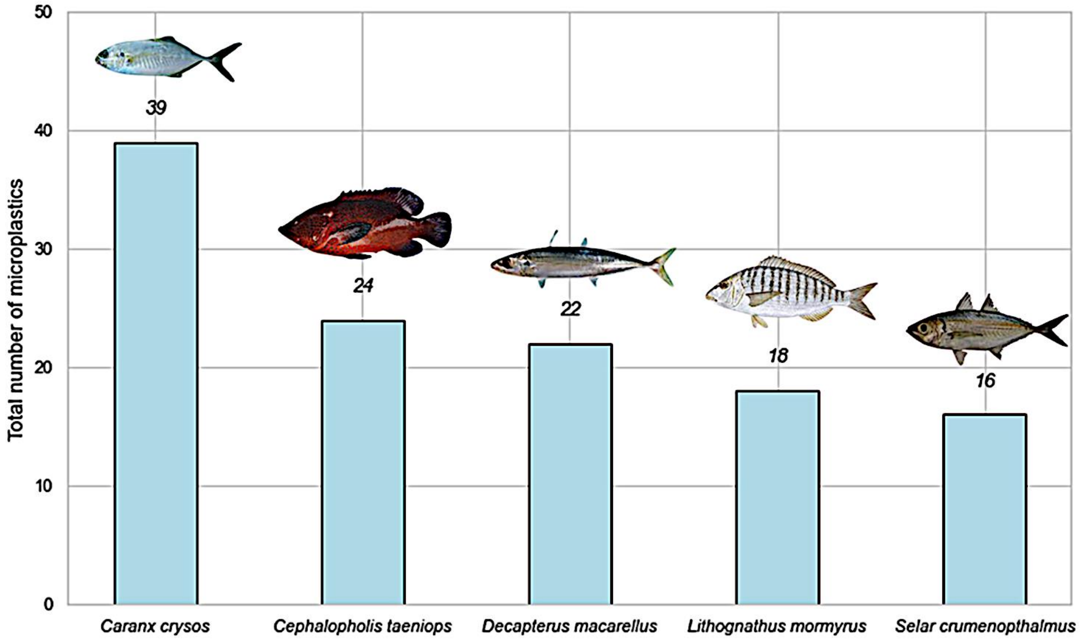

Figure 4: Abundance of microplastics in the fish species (20 per species) sampled near the island of São Vicente, Cabo Verde.

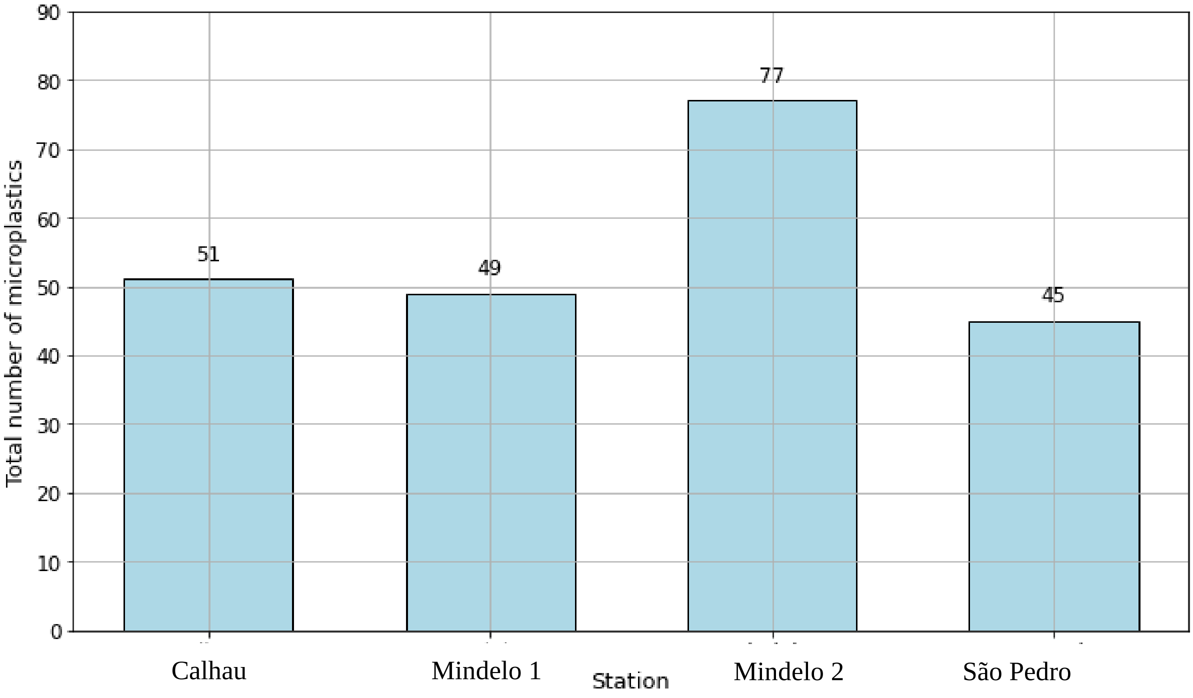

Figure 5: Abundance of microplastics in the field stations sampled (554 m^3 per station) near the island of São Vicente, Cabo. Verde.

4.2 Distribution of microplastics particles in fish

4.2.1 Number of microplastics per fish species

The median number of MP particles ingested by a fish individual was below 1 in all species except for *C. crysos* (Figure 6). The maximum number of MP particles that was detected in one fish individual was 10 (Figure 6).

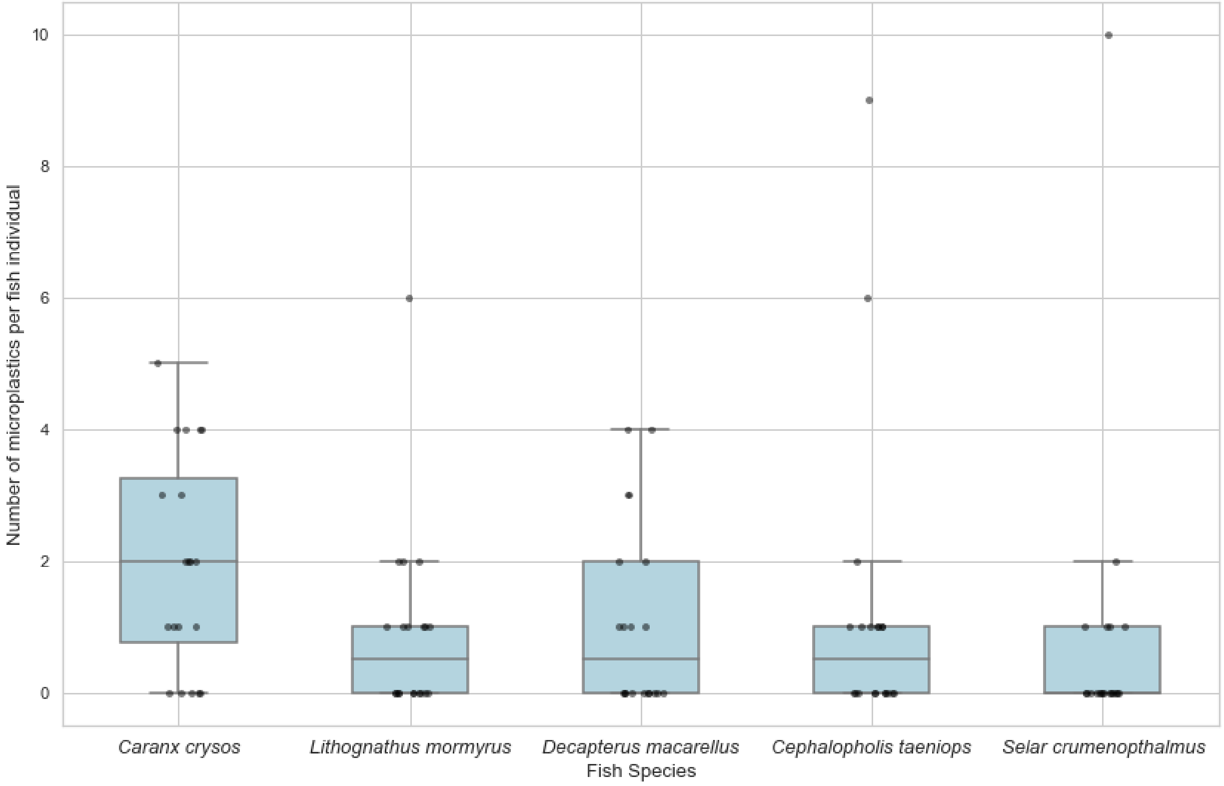

Figure 6: Abundance of microplastics in individuals of the sampled fish species (20 per species). Boxplots show medians, interquartile ranges and non-outlier ranges.

4.2.2 Number of microplastics per particle type

Fibers were the most commonly ingested MP particle type (48) (Figure 7), with *C. taeniops* being the only species in which at least one of all MP types was observed (Figure 8). The majority of MP particle types have a low abundance, with the median abundance being close to 0. However, there are outliers for certain particle types, indicating that they can sporadically occur in higher abundances (Figure 9).

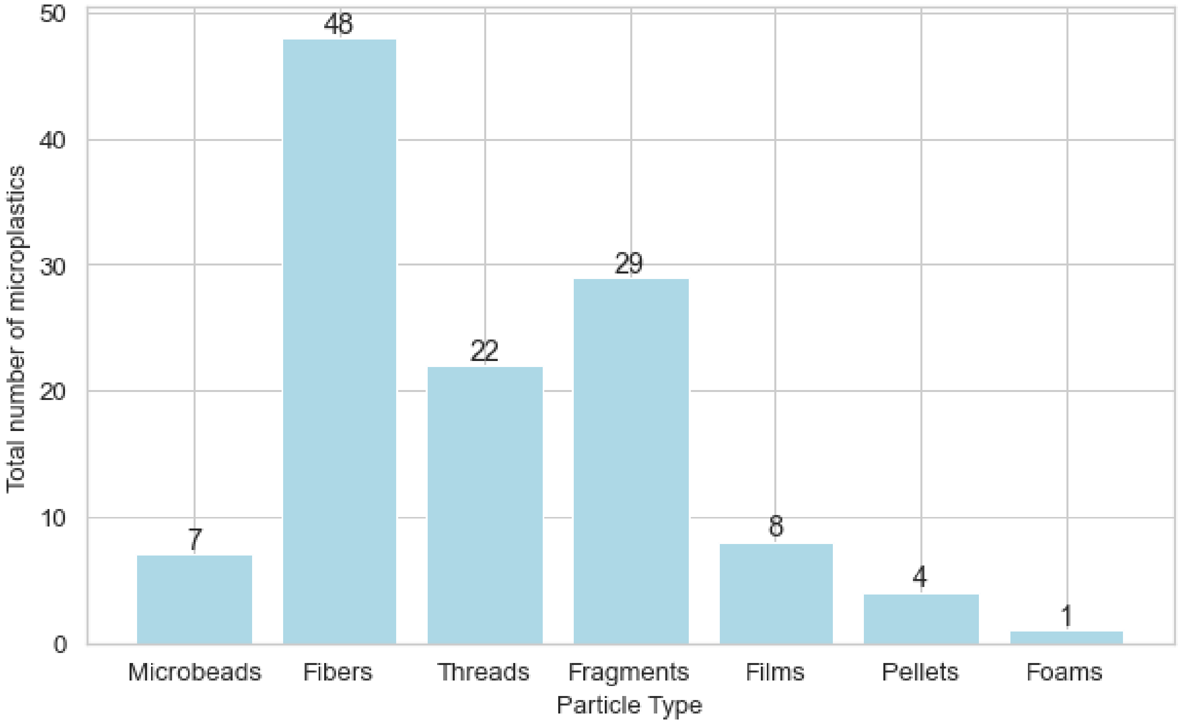

Figure 7: Abundance of different shapes of microplastics in all fish guts (100 individuals, 20 per species) collected near the island of São Vicente, Cabo Verde.

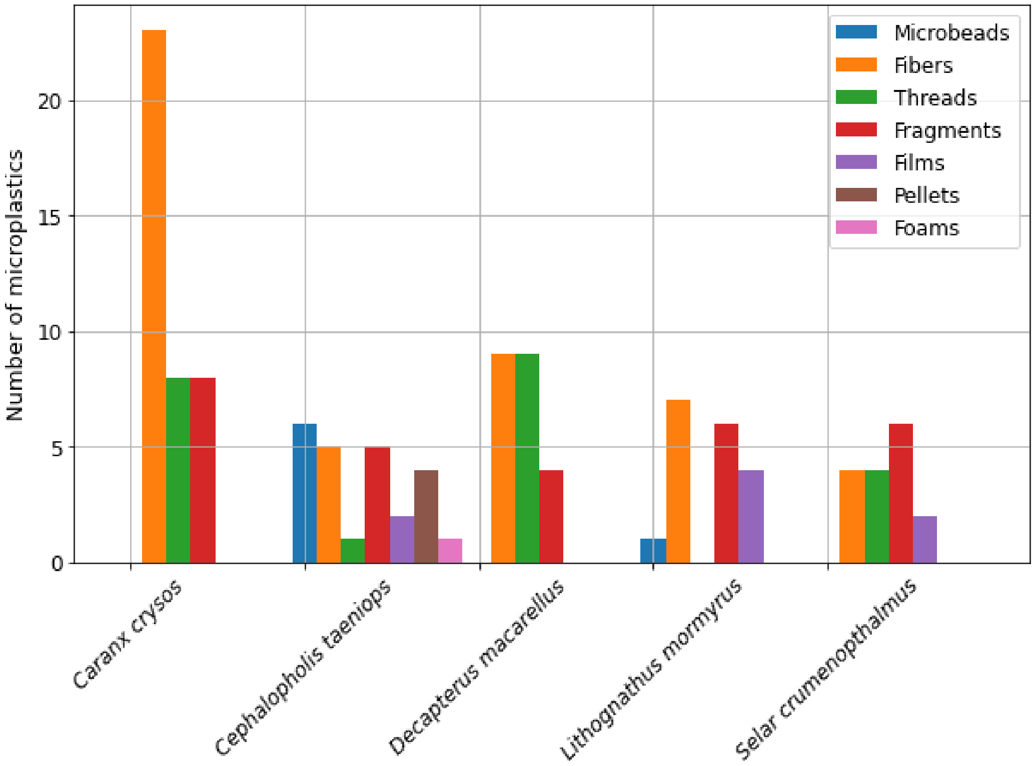

Figure 8: Abundance of microplastics particle types across the fish species (20 per species) sampled near the island of São Vicente, Cabo Verde.

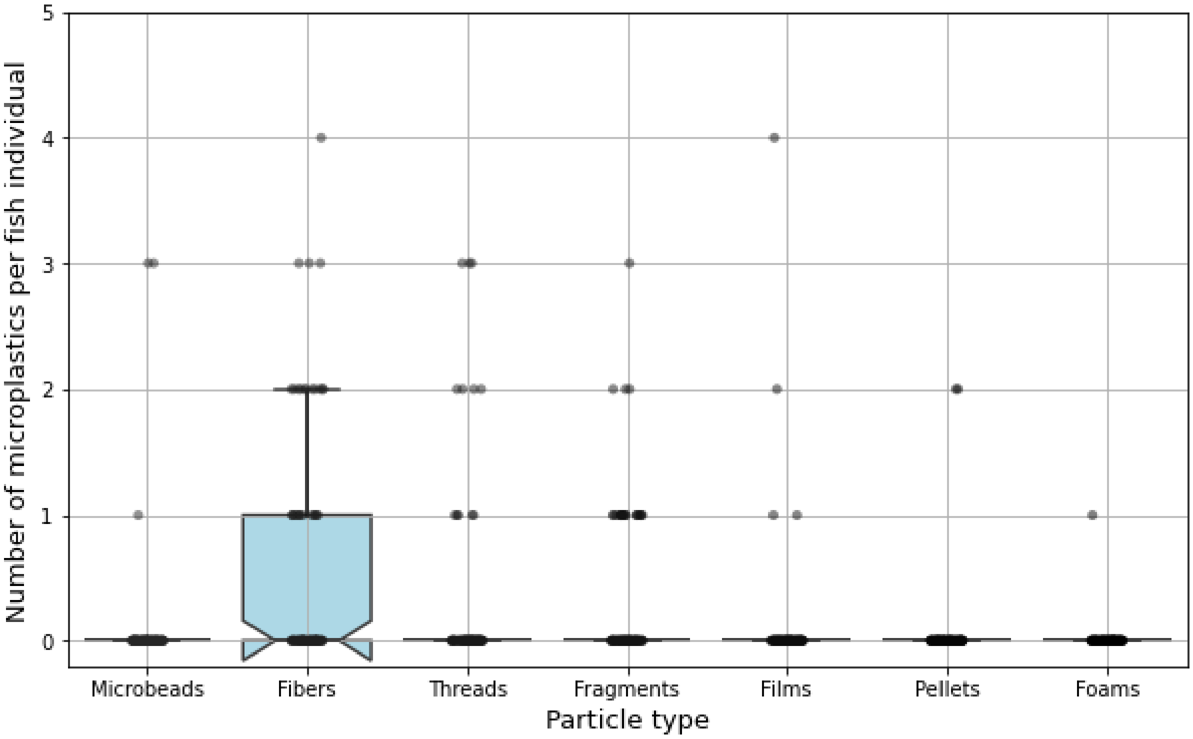

Figure 9: Abundance of different shapes of microplastics found in all fish individuals (100 individuals, 20 per species) collected near the island of São Vicente, Cabo Verde. Boxplots show medians, interquartile ranges and non-outlier ranges.

4.2.3 Number of microplastics per colour

Black was the most common color of MP observed in this study, followed by brown and white. Most colors had a low abundance, with the median abundance being close to 0. However, there were a few outliers for certain colors, suggesting that they sporadically can occur in higher abundances (Figure 10).

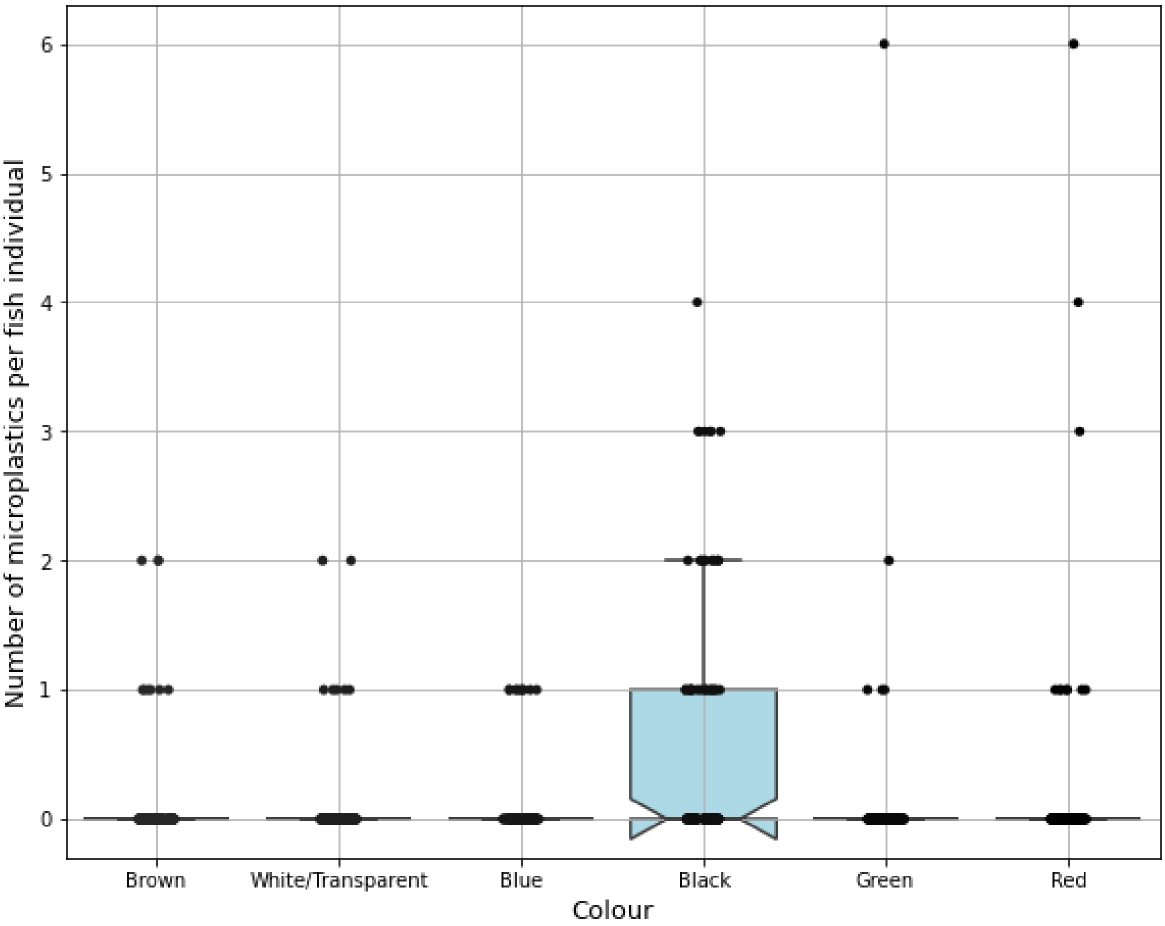

Figure 10: Abundance of different colours of microplastics found in all fish individuals (100 individuals, 20 per species) collected near the island of São Vicente, Cabo Verde. Boxplots show medians, interquartile ranges and non-outlier ranges.

4.3 Abundance of microplastics particles in net tow samples

4.3.1 Number of microplastics across stations

The Mindelo station 2 recorded the highest concentration of MP, with an average of 13 MP particles per 110.8 m^3 of seawater. This abundance was significantly higher than what was observed at the other stations (i.e. São Pedro, Mindelo 1, and Calhau), which showed an average concentration of 5, 5, and 10 MP, respectively (Figure 11).

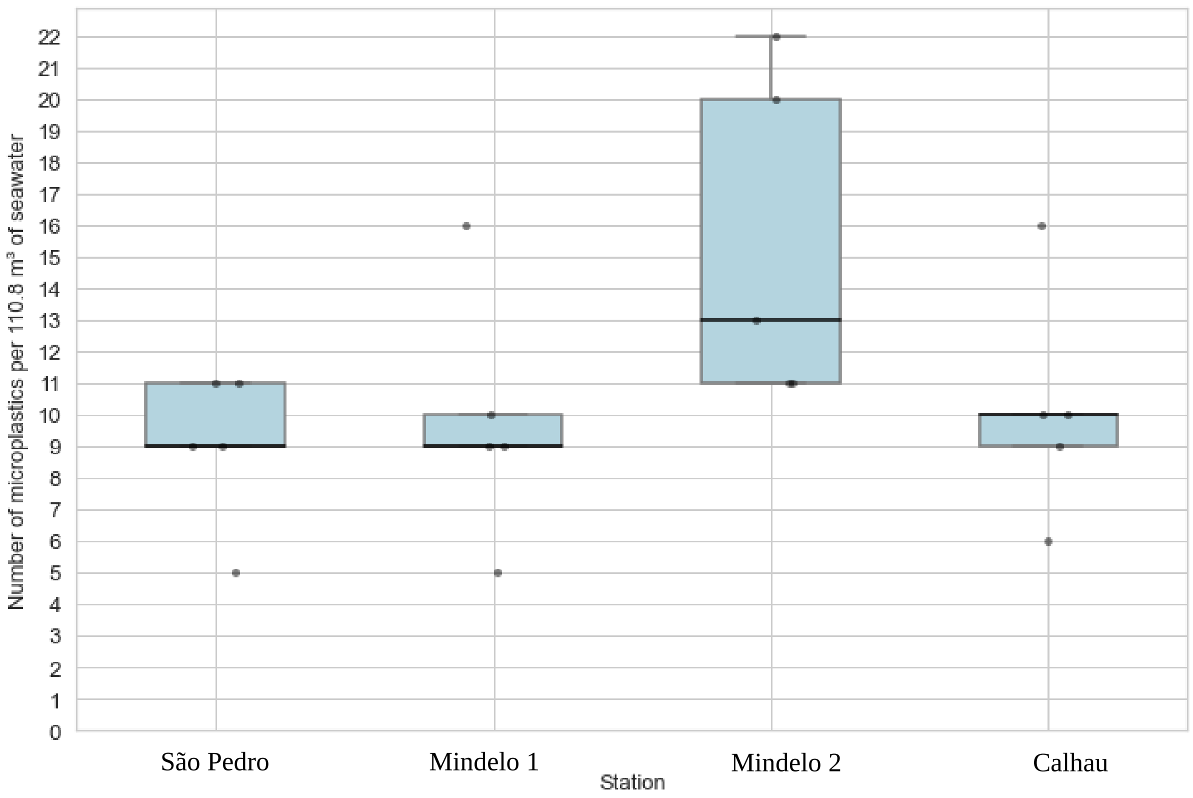

Figure 11: Abundance of microplastics particles in net tow samples (300 µm) taken at four stations along the coastline of São Vicente, Cabo Verde. Boxplots show medians, interquartile ranges and non-outlier ranges.

4.3.2 Abundance of particle types

Fibers, threads, and fragments were the most commonly observed MP particle types across all stations (Figure 12). In all, fibers were the most abundant particle type (Figure 13), with a mean of 6 per station and a maximum of 19. Foam and pellets were not observed in the net tow samples (Figure 14).

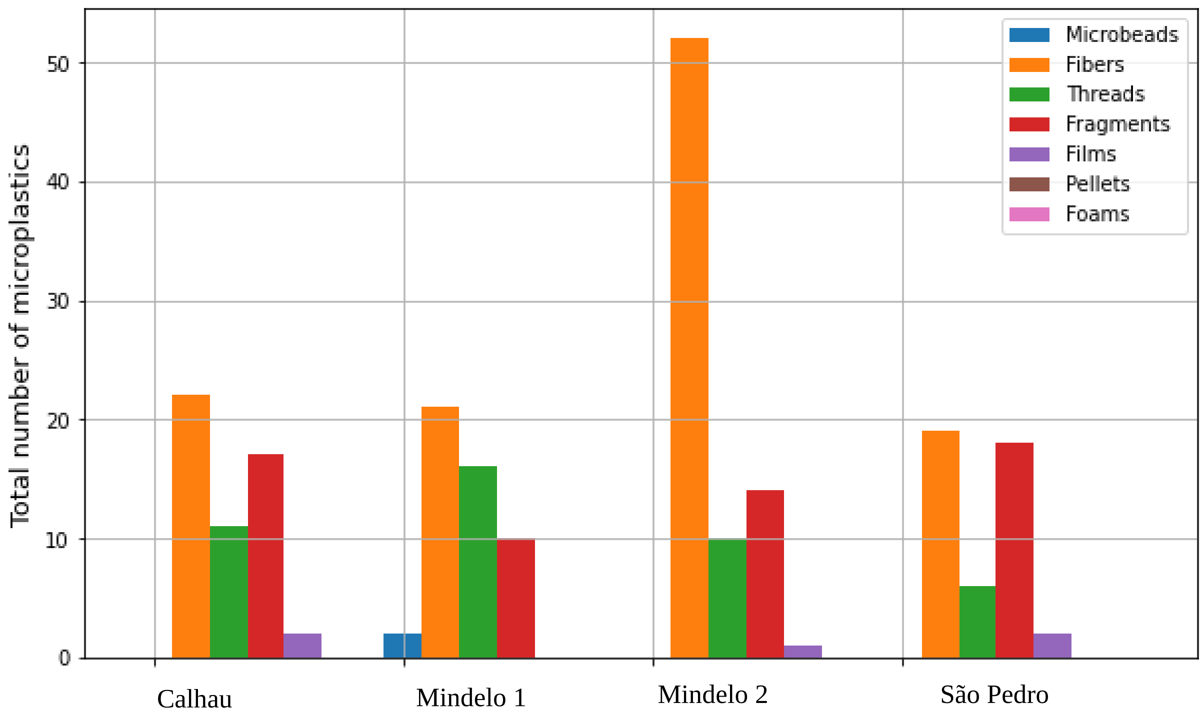

Figure 12: Abundance of different shapes of microplastics particles in net tow samples (300 µm) of 554 m^3 of seawater at each of the four stations along the coastline of São Vicente, Cabo Verde.

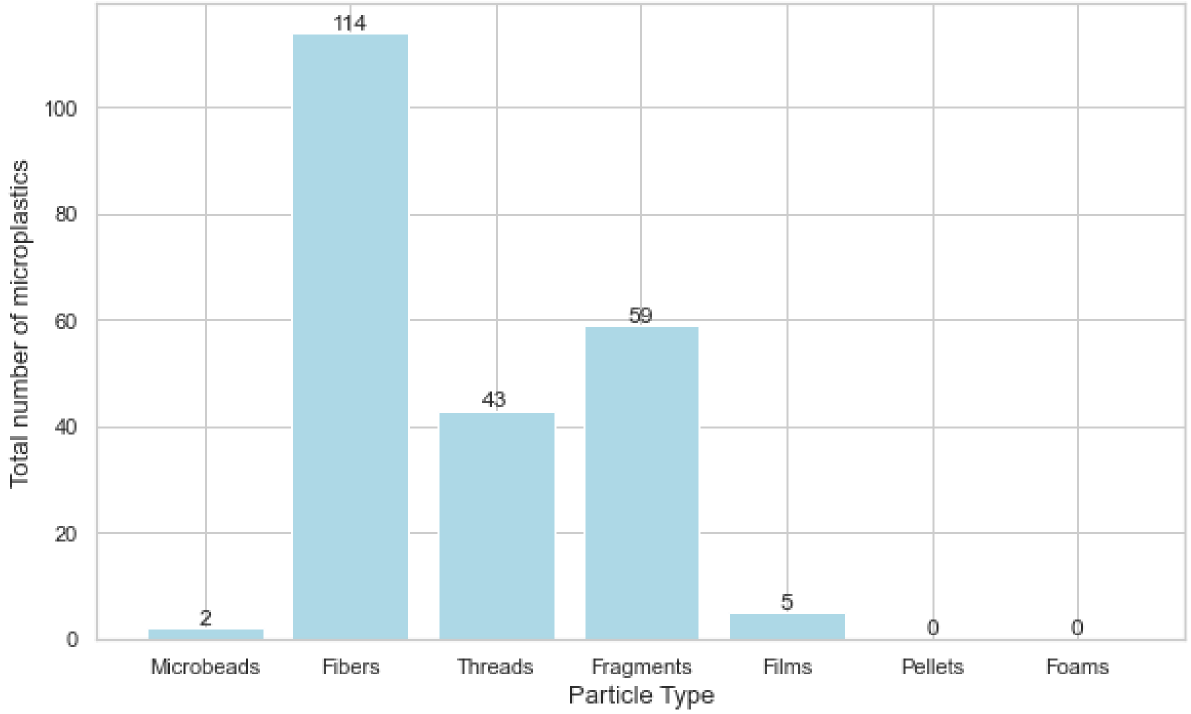

Figure 13: Abundance of different shapes of microplastics in net tow samples (300 µm) of a total of 2216 m^3 of seawater across all four stations along the coastline of São Vicente, Cabo Verde.

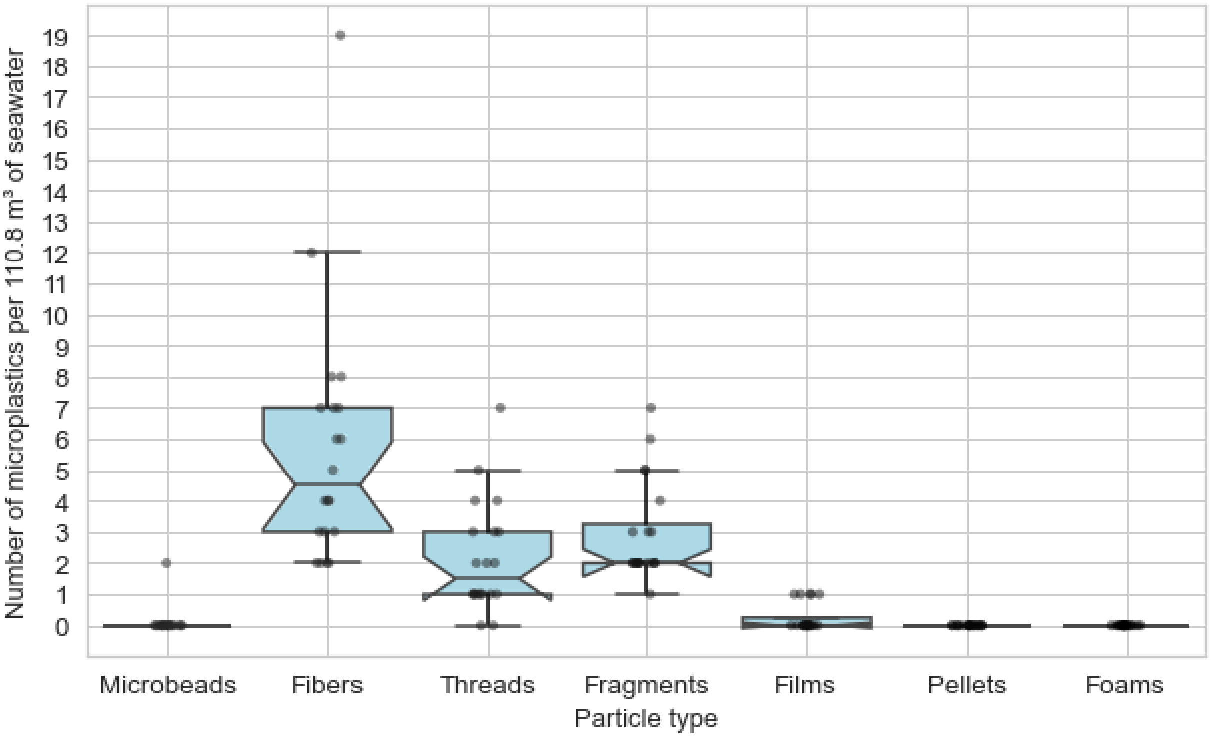

Figure 14: Variation in the distribution of microplastics by shapes in net tow samples (300 µm) of a total of 2216 m^3 of seawater across all four stations along the coastline of São Vicente, Cabo Verde. Boxplots show medians, interquartile ranges and non-outlier ranges.

4.3.3 Number of microplastics per colour

Black colored MP particles were the most observed, followed by brown. Most colors had occurred in similar abundances, with the median abundance being close to 2. However, there were a few outliers for certain colors, suggesting a higher count of MP in those specific color categories (Figure 15).

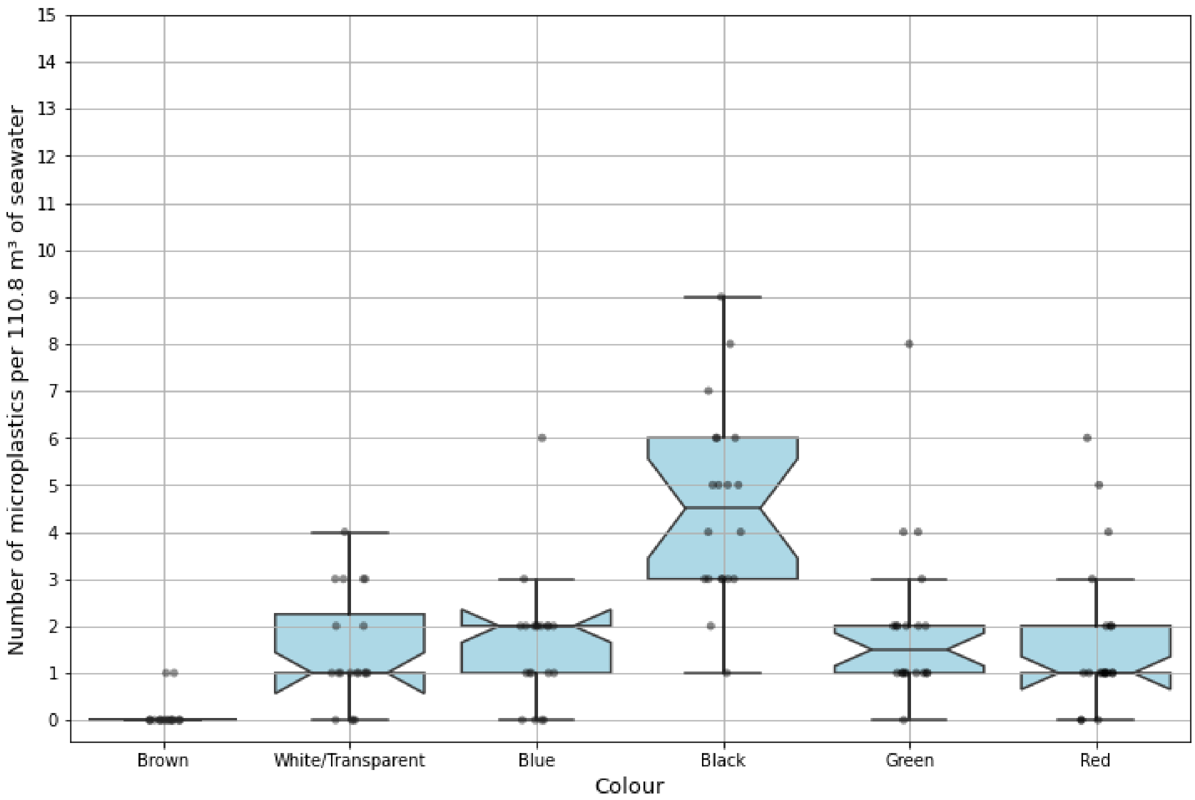

Figure 15: Distribution of microplastics particles by colour in net tow samples (300 µm) of a total of 2216 m^3 of seawater across all four stations along the coastline of São Vicente, Cabo Verde. Boxplots show medians, interquartile ranges and non-outlier ranges.

4.4 Relationship between condition factor and total number of microplastics ingested

A negative correlation was observed between the number of MP found in the gut and the condition factor for *C. crysos* (r-value = -0.26, p-value = 0.260), *C. taeniops* (r-value =-0.18, p-value = 0.458), *L. mormyrus* (r-value = -0.38, p-value = 0.100), *and S. crumenopthalmus* (r-value = -0.38, p-value = 0.097). None of these correlations was significant. However, a positive correlation observed with a r-value = 0.45 and a p-value = 0.047 for *D. macarellus* could indicate a potential relationship between MP ingestion and the condition factor (Figure 16).

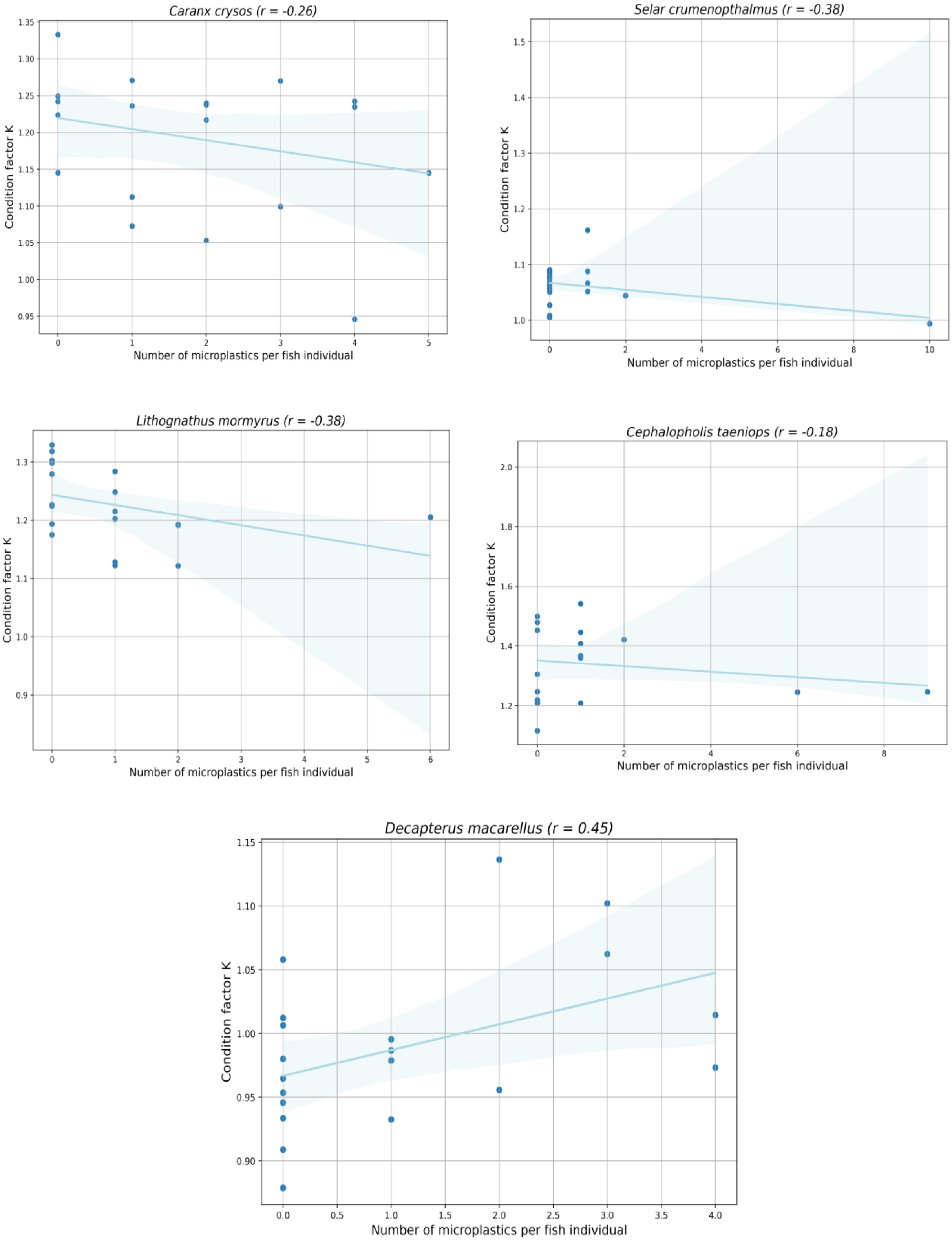

Figure 16: The condition factor was negatively correlated with the number of microplastic particles that was found in for 4 fish species, while it was positively correlated in one.

4.5. Similarity between the microplastics' composition in fish guts

Multidimensional scaling (MDS) was conducted to explore whether the composition of MP (based on shape or colour) differs between fish species. However, this was not the case (Figure 17 and 18). The compositions overlapped, suggesting a lack of preference for a particular MP shape or colour in all the sampled fish species.

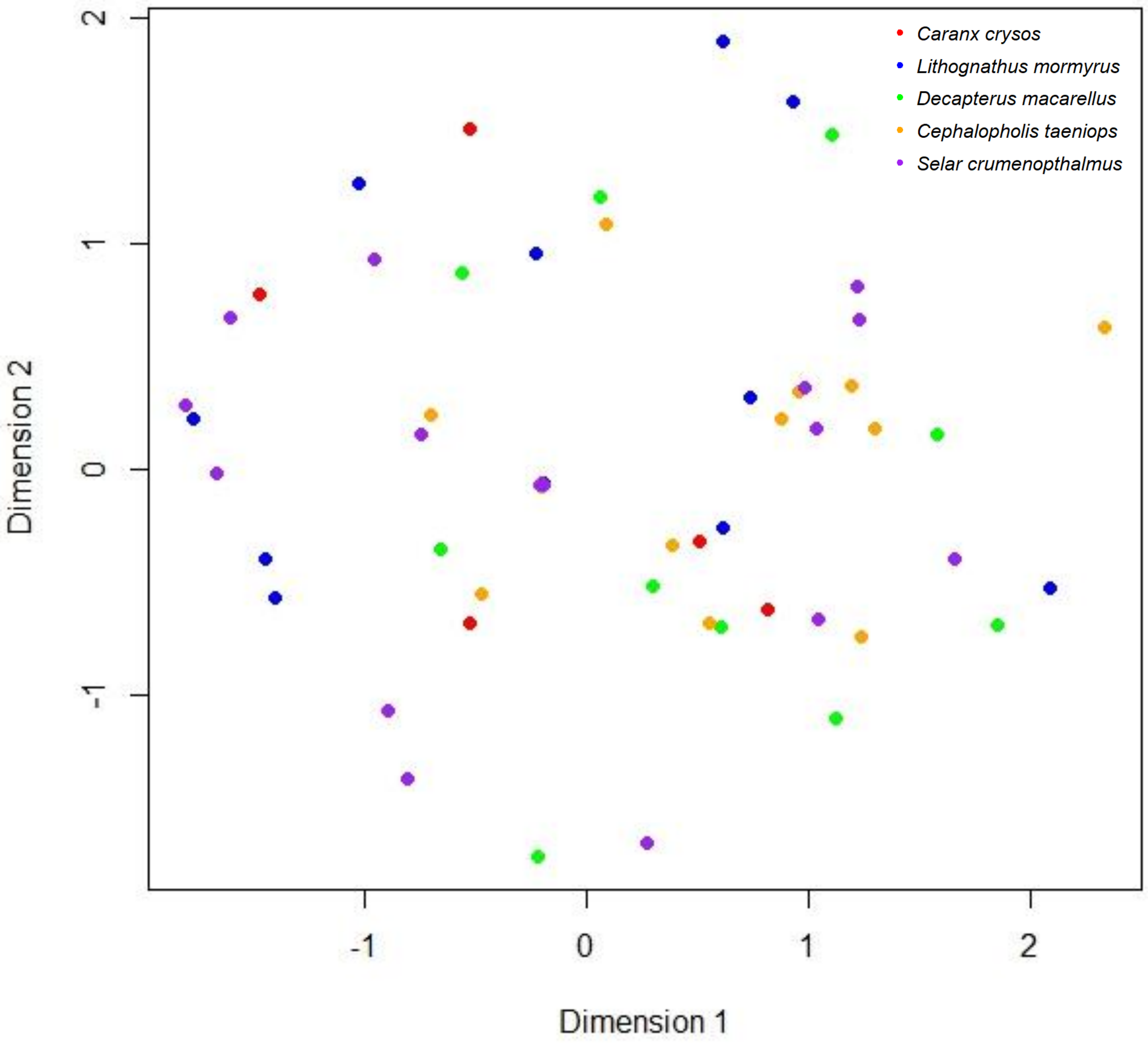

Figure 17: Composition of microplastics based on particle shape found in the guts of the five fish species sampled near the island of São Vicente, Cabo Verde.

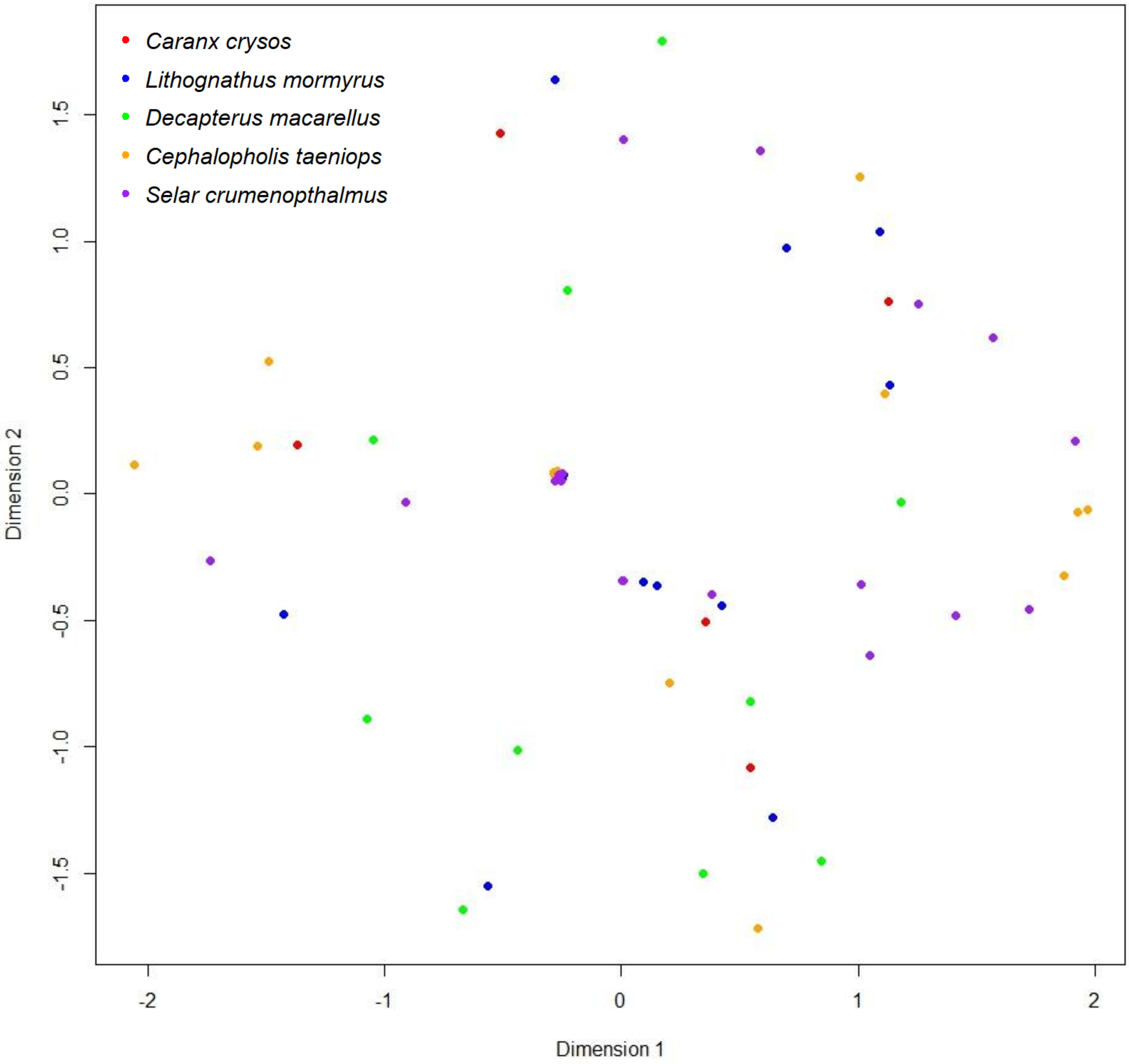

Figure 18: Composition of microplastics based on particle colour found in the guts of the five fish species sampled near the island of São Vicente, Cabo Verde.

4.6 Similarity between the microplastics' composition at the sampling stations

MDS plots were used to analyze the distribution of MP by shape and color across the different stations. There were no discernible patterns or clusters in the composition of MP based on shape or color at any of the stations (Figure 19 and 20). This suggests that the composition of MP was relatively uniform across all the stations.

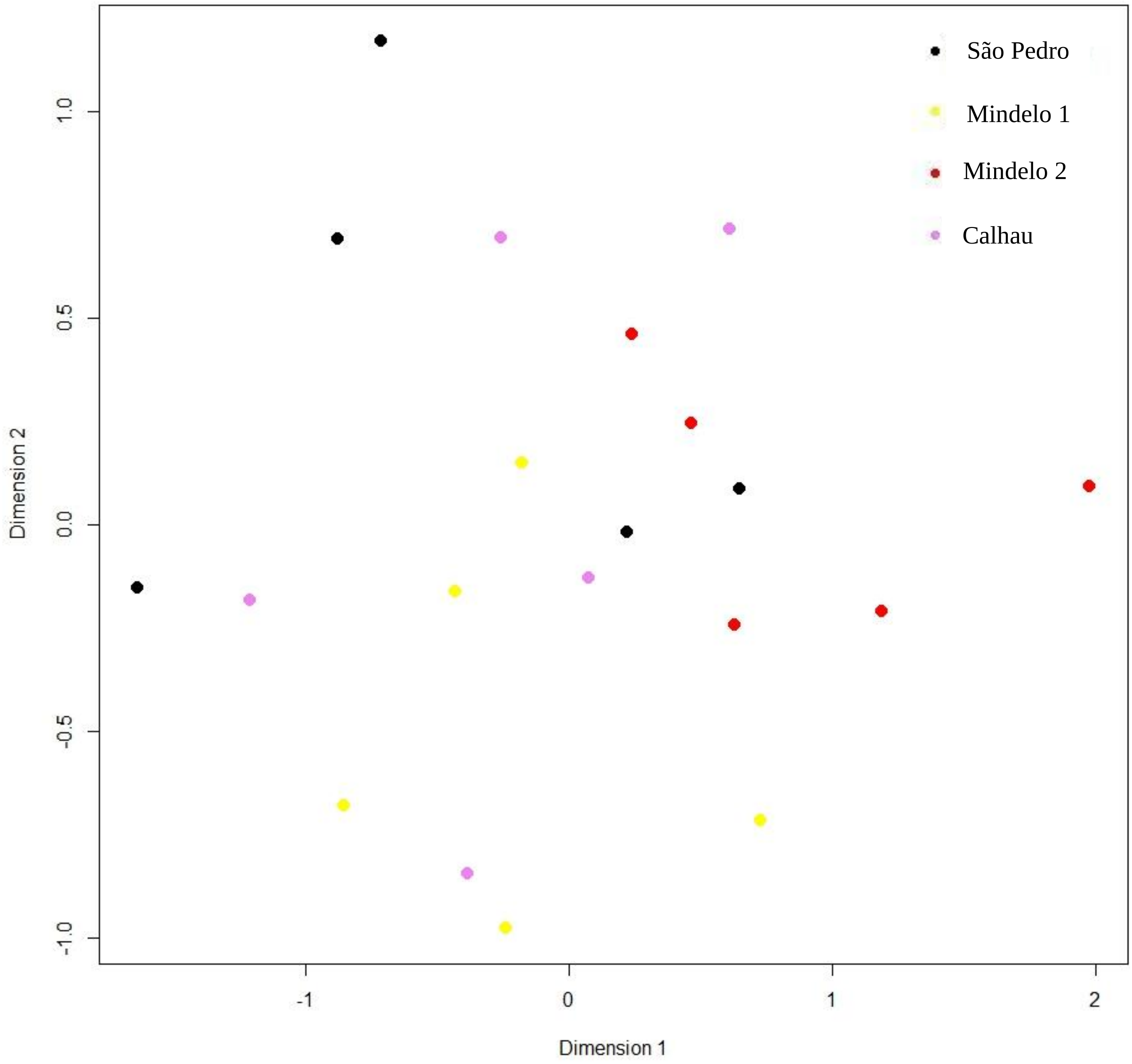

Figure 19: Composition of microplastics based on particle shape found at the different sampling stations near the island of São Vicente, Cabo Verde.

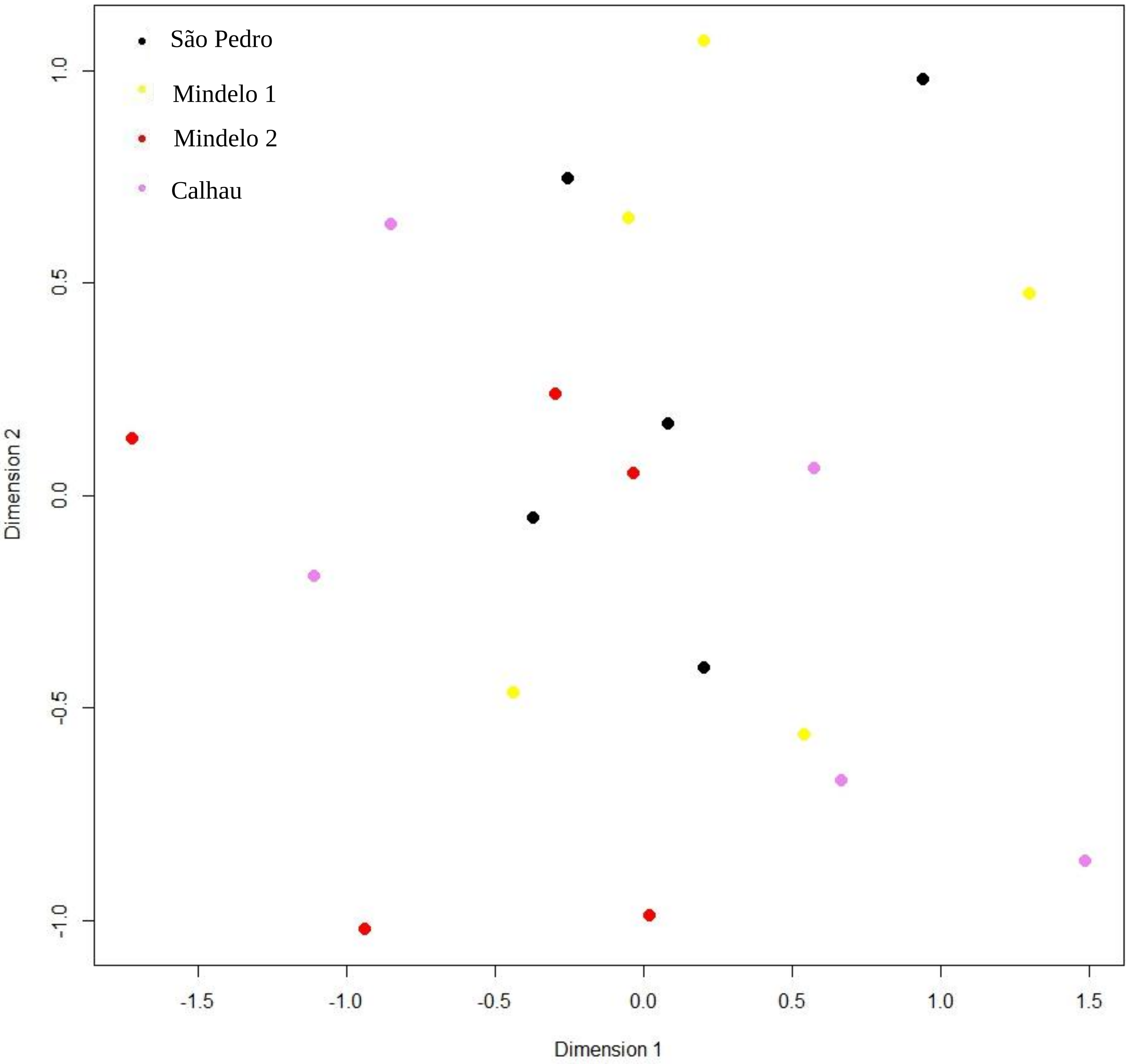

Figure 20: Composition of microplastics based on particle colour found at the different sampling stations near the island of São Vicente, Cabo Verde.

4.7 Relationship between the abundance of microplastic particles in seawater (net tow samples) and in the fish guts

A correlation analysis was conducted on three particle shapes to investigate the relationship between the abundances of MP particles observed in the net tows and those in the fish guts. The data on MP from fish was smoothed using a moving average filter to ensure that it was the same length as the data from water before the correlation calculation was performed. Results showed that fragments and fibers in sea water and fish guts correlated positively with a correlation coefficient of 0.36 (p-value = 0.116) and 0.49 (p-value = 0.029) respectively, while thread correlated negatively with a correlation coefficient of -0.23 (p-value = 0.333) (Figure 21).

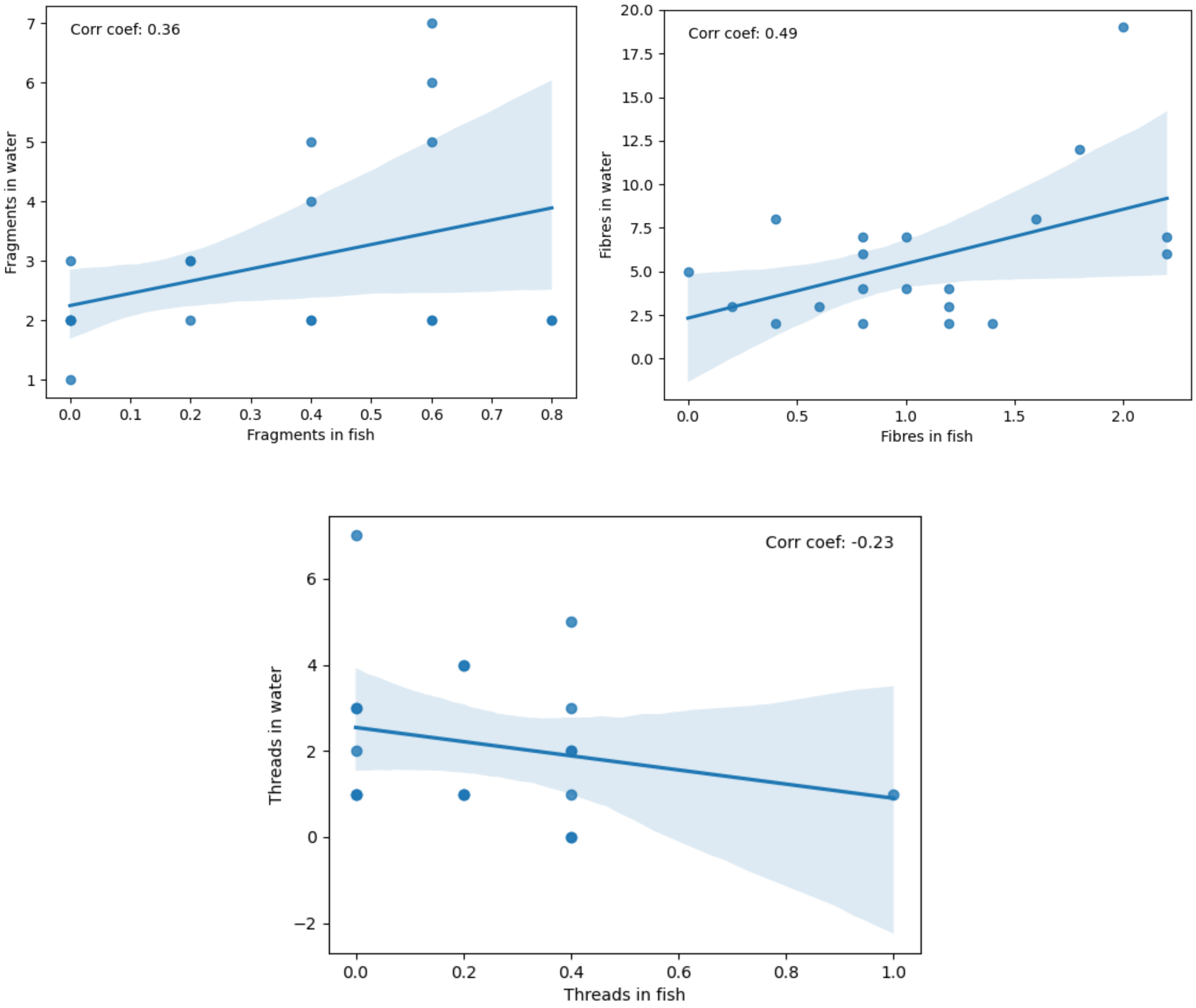

Figure 21: Correlation between microplastics particles types in seawater and fish gut samples.

4.8 Polymer analysis of microplastic particles

Of the total 341 MP particles detected in this study, 25 particles with a minimum length of 300 µm were selected for polymer analysis using the hyperspectral imaging (HSI) system, which accounted for 7.3% of the total MP observed. From these 25 particles, the HSI system identified 6 as non-plastic material. The remaining 19 particles were classified into two polymer types: polyethylene (PE) and polypropylene (PP). Among the 19 particles, 13 were identified as PE with an average confidence level of 58%, while the remaining 6 were identified as PP with a mean confidence level of 37% (Figure 22 and Table 2).

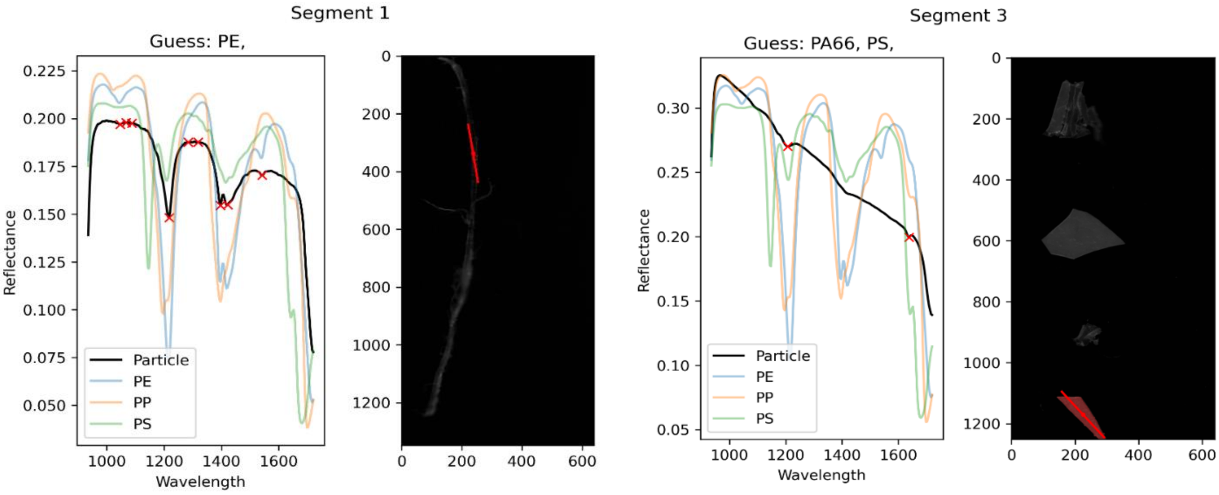

Figure 22: Identified spectral by HSI camera prior to final classification.

Table 2: Microplastic particles that were analyzed with the HSI camera.

Folder	prediction	non_plastic	PVC	PS	PP	PMMA	PET	PE	PC	PA66
GV01	PE	0.12	0.05	0	0.09	0.03	0	0.71	0	0
LM3	PE	0.24	0.11	0.04	0.19	0.04	0	0.3	0.02	0.06
LM16	non_plastic	0.51	0.05	0.03	0.16	0	0.01	0.24	0	0
CC-20	PP	0.14	0.05	0.07	0.33	0.01	0.02	0.25	0.04	0.09
CC-20	PE	0.41	0	0.02	0.03	0.01	0.01	0.43	0.01	0.08
CC-20	non_plastic	0.8	0.06	0	0.08	0	0	0.06	0	0
DM03	PP	0.14	0.01	0.15	0.41	0.04	0.03	0.19	0.01	0.02
DM15	PP	0.13	0.02	0.03	0.5	0.07	0.03	0.2	0	0.02
DM19	PP	0.09	0.04	0.07	0.42	0.01	0.07	0.22	0	0.08
Min_SP	PE	0.07	0	0	0.01	0.01	0.01	0.85	0.02	0.03
Min_SP	PE	0.2	0	0.02	0	0.02	0	0.76	0	0

5. Discussion

5.1 Microplastic ingestion by fish species

The ingestion of microplastics (MP) by fish is a well-known phenomenon resulting from their mistaken identification of these particles as food (Cole & Galloway, 2015). In this study, MP particles were found in the gastrointestinal tracts of 51% of the analyzed fish individuals. The abundance of MP varied among fish species, with *Caranx crysos* showing the highest (33%) and, *Selar crumenopthalmus* with the lowest prevalence of MP (13.5%). Furthermore, the composition of MP in fish guts with regard to shape and color showed random variability among species.

Comparable to studies that investigated the occurrence of MP in the gut of fishes (e.g. Harikrishnan et al., 2022; Hastuti et al., 2019; Jiang et al., 2023; Piskuła & Astel, 2023), fibers were found to be the most abundant MP type representing 40% of the entire MP identified. Fibers are small, elongated fragments of synthetic materials commonly encountered in the marine environment, arising from the degradation of larger plastic particles (Browne et al., 2011; Hidalgo-Ruz et al., 2012). Their ubiquity in marine ecosystems stems from their extensive utilization in various consumer goods, such as clothing, carpets, and fishing nets, as well as their release during laundering of synthetic textiles and wastewater discharge (Graca et al., 2017; Tang et al., 2018). Owing to their diminutive size, fibers often resemble natural prey items, such as plankton organisms, rendering them prone to inadvertent ingestion by fishes especially filter-feeding species and those consuming suspended particulate matter. Additionally, their buoyant characteristics enable them to remain afloat on the water's surface or remain suspended within the water column, enhancing the likelihood of incidental interactions with fish during feeding activities (Santini et al., 2022).

Fragments accounted for 18.5% of the total identified MP particles, ranking as the second most prevalent MP type across all fish species. However, higher fragment abundances have been reported in other studies. For instance, Pereira et al. (2020) observed fragments as the most abundant MP type in the guts of pelagic fishes from the Azores archipelago, followed by fibers. Similarly, investigations of the guts of fish from the coastal waters of Cabera archipelago Maritime Terrestrial National Park (Alomar et al., 2016) and the oceanic islands of the Western Tropical Atlantic oceans (Ivar do Sul et al., 2014) showed higher occurrences of fragments compared to fibers. In contrast, Herrera et al. (2019) found fibers to be the most prevalent MP type, followed by fragments, in the gut of fishes from the Canary Islands, which aligns with the trend observed in this study.

This study demonstrates that fibers and fragments are the dominant types of MP present in the marine environment, which aligns with findings from previous research. The prevalence of these MP types can be attributed to their widespread use in consumer products, such as textiles and packaging materials, as well as fishing gears, which subsequently contribute to their introduction into the environment through inadequate waste management practices and fishing activities (Ferreira et al., 2021).

The results of this study also confirmed our anticipation that *C. crysos*, with its benthopelagic nature, would exhibit a higher MP load, as it has the opportunity to feed near the surface, throughout the water column, and at the bottom.

Additionally, previous studies by Kristanti et al. (2022) and Phaksopa et al. (2021), reported higher MP abundance in the guts of pelagic fish species compared to demersal species. This finding might be attributed to the tendency of MP to float on the ocean's surface and be suspended within the water column, making them more accessible to pelagic species. However, this study revealed a higher abundance of MP in the guts of demersal fish species (*C. taeniops* and *L. mormyrus*) than in the pelagic ones (*D. macarellus* and *S. crumenopthalmus*). Various factors, such as distinct feeding behaviors, foraging preferences, and the availability and types of MP present in the respective habitats of these fish species could influence this deviation from the anticipated trend. The complex interactions between fish species and MP ingestion warrant further investigation to comprehensively understand the underlying mechanisms driving the observed variations in MP abundance among different fish species. Notably, the findings of this study align with those of Murphy et al. (2017) and Bellas et al. (2016), which also reported higher MP abundance in demersal fish species. Thus, there seem to be no predominant picture with regards to the ingestion of MP by pelagic or demersal fish species. This indicates the need for a more comprehensive and region-specific understanding of MP ingestion patterns in different marine ecosystems.

It is also worth noting that, the demersal fishes covered in this study, were found to ingest microbeads, which are more likely to sink through the water column towards the sea bottom because of their small size and relatively high density (positive buoyancy property) compared to the surrounding water. This finding aligns with several studies (Adika et al., 2020; Mahu et al., 2023) that recorded higher occurrence of microbeads in demersal than in pelagic fishes.

A study conducted by Fernandes (2019) confirmed MP ingestion by fish species (*Sparisoma cretense*, *Cephalopholis taeniops* and *Diplodus prayensis*) that were caught near Santa Luzia, an uninhabited island in Cabo Verde. The authors found a relatively higher MP abundance (94.5% of all the individuals sampled contained MP) compared to the findings from this study. The differences in MP abundance between Santa Luzia and São Vicente can be attributed to the more polluted waters around Santa Luzia, resulting from the input of plastics by the Canary Current (Cardoso & Caldeira, 2021; Fernandes, 2019). Furthermore, the Canary Current impacts both locations, but its influence is more significant on Santa Luzia, leading to higher MP abundances there. Additionally, Santa Luzia is closer to the northwestern African coast, which has been identified as a potential source of land-based particles, including MP, into the Cabo Verde archipelago (Cardoso & Caldeira, 2021). This proximity could explain the higher MP abundances observed in the waters of Santa Luzia.

Consistent with prior research by Ory et al. (2018), this study also revealed a predominant ingestion of black-colored MP particles by fish. Moreover, this higher abundance of black MP particles can be attributed to their availability primarily in the form of fibers. Consequently, the observed prevalence of black-colored MP particles corresponds to the prevalence of fibers, which ultimately explains why fibers were the most commonly ingested type of MP. Likewise, another study from Bellas et al., (2016) revealed the occurrence of black coloured MP particles in demersal fishes sampled along the Spanish Atlantic and Mediterranean coast, with fibers also being the most occurring MP particles by shape.

Additionally, the lack of distinct clustering of MP particles with regard to shape or color across the fish species, as observed in the Multidimensional Scaling (MDS) analysis, suggests that fish species did not exhibit selectivity in MP ingestion, ingesting whatever was present in their environment. However, it is essential to acknowledge that the sampling conducted in this study represented only a snapshot of MP presence at a specific moment in time, and the residence time of MP within the fish is likely to be relatively short, possibly lasting only several days. Consequently, the observed pattern of MP ingestion may not reflect a permanent situation or long-term accumulation within the fish species.

Regardless of the MP ingestion by fish observed in this study, MP ingestion has been associated with detrimental effects on fish, including a decrease in their growth (Besseling et al., 2013; Jovanović, 2017; Ogonowski et al., 2016). In this study, all individuals of *C. crysos, C. taeniops, L. mormyrus* and *S. crumenopthalmus* were found to be in good condition, indicating that MP ingestion, given that it occurred to the same extent observed in this study for a longer time period did not have a significant impact on these fish species. However, individuals of *D. macarellus* exhibited poor growth conditions, and a positive correlation (p-value 0.047) was observed between the condition factor and the MP particles ingested, suggesting a potential impact of MP ingestion on their growth. It is important to note that *D. macarellus* did not record the highest number of MP ingested, and not all samples from this species ingested MP. Therefore, the ingestion of MP alone cannot be solely responsible for their poor growth conditions, as other factors may contribute to their condition.

In a study conducted by Alomar et al. (2016) it was observed that ingestion of MP by red mullet (*Mullus surmuletus*) did not result in oxidative stress or cellular damage in the liver. However, a moderate elevation in the activity of glutathione S-transferase (GST) was detected, suggesting a potential activation of detoxification mechanisms. Another study from the North Sea also did not find a significant correlation between MP ingestion and the overall condition of different fish species (Foekema et al., 2013). These findings highlight the potentially complex nature of the relationship between MP ingestion and fish health and why it is important to consider other environmental factors including habitat quality and food availability (Parrish & Mallicoate, 1995).

While fish guts' contents provide valuable insights into the presence of MP in the marine environment and their ingestion by fish, it is essential to recognize that the examination of gut contents represents only a snapshot of the MP pollution levels and the potential feeding preferences of the fish at a specific moment in time. MP ingestion may vary depending on various factors, such as the availability and concentration of MP in the water, the fish's feeding behavior, and the type of MP present. Moreover, fish may selectively ingest certain types of MP or avoid ingesting others, leading to a bias in the observed ingestion patterns. Additionally, gut content analysis does not provide a comprehensive picture of the overall health of the fish or the long-term impact of MP ingestion on their well-being. Other physiological and ecological factors can influence the health of fish, and MP may interact with these factors in complex ways. Therefore, while gut content analysis offers valuable data, it is crucial to complement this information with other research methods to gain a more comprehensive understanding of the effects of MP on fish and the marine environment (Adika et al., 2020).

It is therefore recommended that future studies consider and explore these additional factors to gain a comprehensive understanding of the relationship between MP and fish growth.

5.2 Impacts of microplastic ingestion on the marketability of fish products

As current research endeavors focus on examining the potential impacts of MP ingestion on the well-being of fish and humans (Jovanović, 2017; von Moos et al., 2012), it becomes essential to carefully consider how the ingestion of MP by fish could affect the marketability of these fish. These implications primarily revolve around concerns regarding food safety and consumer perception.

When fish ingest MP, there is a potential for harmful substances associated with MP to accumulate in the tissues of the fish (von Moos et al., 2012). These substances can include chemicals and pollutants that may be present in the MP or adhere to their surfaces (von Moos et al., 2012). Consequently, consuming fish with accumulated MP may pose implications for human health.

From a food safety perspective, the presence of MP and associated contaminants raises concerns regarding potential health risks for consumers (Wright & Kelly, 2017). Regulatory agencies and consumers are increasingly concerned about the presence of MP in food products and their potential impact on human health. Consequently, fish with high levels of MP ingestion may face challenges in meeting food safety standards and regulations, affecting their marketability (Rivera-Garibay et al., 2023).

Moreover, consumer perception plays a significant role in the marketability of fish. With growing awareness of MP pollution, consumers are becoming more conscious of the environmental impact on their food choices. The presence of MP in fish can be perceived as a sign of environmental contamination and poor quality, leading to consumer hesitancy in purchasing and consuming these products (Rivera-Garibay et al., 2023). Fisheries and seafood industries may face reputational damage if their products are associated with high levels of MP contamination.

Additionally, the guts of fish are not always removed during culinary preparation, such as frying or smoking (Mahu et al., 2023), and this may have implications for human health if MP are present in the gut contents. If the public becomes aware of this situation, the marketability of these fish products may decrease. It is also worth noting that fish gut contents are sometimes used to formulate fish feed for aquaculture (Mahu et al., 2023). This can contribute to the transfer of MP to farmed fish. If the public becomes aware of the potential dangers associated with MP in farmed fish, the marketability of these products may also be adversely affected.

In Cabo Verde, it is commonly observed at fish markets that fishmongers remove the gut contents of fish before selling them. While this practice is commendable, there may still be a secondary form of MP pollution on the fish tissues if they are not thoroughly washed. Furthermore, the gut contents are sometimes improperly disposed of by being thrown back into the nearby sea. It is often observed that juvenile fishes surround those areas, feeding on the thrown guts content into the sea. This activity can result in further MP pollution in other fish, potentially affecting the wider food web and exacerbating the menace of MP pollution.

5.3 Occurrence and distribution pattern of microplastics across stations

The presence of MP particles in seawater is also a well-documented global issue, with fibers recorded as being one of the most commonly observed shape (Mu et al., 2019; Wakkaf et al., 2020; Wang et al., 2020). In this study, a total of 222 MP particles were identified across the sampled stations.

These particles enter coastal waters and open oceans through various sources, including inadequate waste management and littering, industrial activities such as manufacturing and waste disposal, urban runoff from streets and storm drains, discharge from wastewater treatment plants, atmospheric deposition, and maritime activities like fishing and shipping (Browne, 2015).

The composition of MP by shape in this study followed a consistent trend, with fibers, fragments, and threads being the most prevalent types across all stations. This finding aligns with previous studies highlighting the abundance of fibers in the marine environment (Andrady, 2011; Kiliç et al., 2022). The high abundance of fibers observed in this study could be attributed to factors such as the breakdown of fishing gears over time and the shedding of MP from textiles during washing machine cycles as well as the breakdown of other consumer products that are released into the environment (Adika et al., 2020; McCormick et al., 2014).

Interestingly, very low numbers of pellets (0) and microbeads (2) were observed across all stations. This could be attributed to their higher tendency to sink to the benthic environment due to their lower surface area to volume ratios, compared to fibers, threads, and fragments, which have greater surface area to volume ratios and are more likely to float on or within the water column (L. Li et al., 2018). Another reason for their observed low abundances can be attributed to the absence of potential sources, including factories and sewage discharge, plastic processing plants, and offloads from shipping terminals or ports (Norén and Ekendahl, 2009). While Cabo Verde has a significant shipping trade, it does not involve the transportation of pellets and microbeads in its waters, thus limiting the input of these particles into the marine environment (*Trade Profile - Cape Verde - International Trade Portal*, n.d.). This absence of specific sources is likely a contributing factor to the low levels of pellets and microbeads observed in this study. This result aligns with studies from Ivar do Sul et al. (2014) who also attributed the low levels of pellets observed around the oceanic islands of the Western Tropical Atlantic Oceans to the absence of primary sources for this kind of MP.

The composition of MP by color (brown, white/transparent, red, green, blue, and black) was found to be similar across all stations. This finding is consistent with other studies on the common colors of MP found in the marine environment (Hidalgo-Ruz et al., 2012; Pan et al., 2022; W. Zhang et al., 2017). Other studies however, found white and brown colored MP to be more abundant than differently coloured MP particles in marine environments as a result of weathering and the generation of chromophore products from the oxidation of MP as they are exposed to sunlight (Abaroa-Pérez et al., 2022; Zhao et al., 2022). Interestingly, this study revealed black colored MP particles as the most abundant type. The prevalence of black MP suggests that these particles may be relatively new and have not undergone significant weathering or discoloration. Potential sources of these black-colored MP include the gradual breakdown of car tires over time, which releases particles into the marine environment, as well as the shedding of black synthetic textiles from washing machines and fishing nets. However, it is important to acknowledge that this study did not extensively investigate other factors such as water temperature, salinity and currents, among other environmental characteristics that could influence the distribution and transport of MP in marine ecosystems (Auta et al., 2017).

The abundance of MP particles was notably higher at the Mindelo 2 station than at the others. This observation could be attributed to the proximity of the station to the ports (Porto Grande), where port activities and the passage of ships and vessels could introduce MP particles into the surrounding region. Interestingly, at Mindelo 1 station, which is located near a very popular and crowded beach (Laginha Beach), levels of MP particles were not particularly high. This could be attributed to the complex interplay of tides, winds, currents, and wave actions that may redistribute MP away from the beach (Ballent et al., 2012; Brander et al., 2020; Cardoso & Caldeira, 2021; Cole & Galloway, 2015; D’Iglio et al., 2022; Fred-Ahmadu et al., 2020; Hidalgo-Ruz et al., 2012; Schmidt et al., 2018).

Also, as observed from the MDS analysis, there was no form of MP clustering either by shape or color across the 4 stations. This suggests that there are no factors that determine a certain spatial distribution of differently shaped or colored MP.

Comparing the findings of net tow samples of seawater to those of fish samples, a clear similarity emerges in the composition of MP by shape and color. In both water and fish samples, fibers, fragments, and threads were observed to be the most abundant types, in that order. This consistency suggests that the fish's ingested MP may reflect what was available in their surrounding environment.

5.4 Polymer identification

Polymer identification of selected MP particles using the hyperspectral imaging (HSI) camera revealed the presence of two polymer groups: polyethylene (PE) and polypropylene (PP). Among the analyzed samples, 52% were identified as PE, while 24% were identified as PP. Additionally, six particles were found to be non-plastics. These findings are consistent with previous observations of commonly encountered polymers in marine environments and fish species (Al Nabhani et al., 2022; Alomar et al., 2016; Andrady, 2011b; Pereira et al., 2020), as these polymers are more widely produced, used, and disposed of into the environment and can to stay afloat on the seawater surface because of their lightweight compared to seawater. This supports the earlier argument regarding the potential contribution of littering, mismanaged waste, shedding of MP from textiles during washing with washing machines, and the fragmentation of fishing gears to the issue of MP pollution.

It is essential to acknowledge that only 25 MP particles were analyzed in this study, representing only a small fraction of the total MP detected. This limitation arises from the constraints of the HSI system utilized, which is unable to analyze black and white colored particles and has a minimum size requirement of 300 µm for MP. Most of the MP identified were black in color and the sizes were less than the minimum 300 µm. All identified MP particles were confirmed using the protocols by Lusher et al. (2017), including the hot needle approach. However, certain MP particles were misidentified as non-plastic in the results obtained from the HSI. This misclassification may be due to obstructions on these particles, hindering accurate infrared light reflection during analysis. Consequently, the HSI failed to obtain precise spectral information from these obstructed particles, incorrectly categorizing them as non-plastic.

Nonetheless, this study provides valuable information about the abundance and composition of MP and potential MP sources such as fishing gears and inadequate waste management by individuals leading to pollution in the marine environment around the São Vicente Island of Cabo Verde.

6. Conclusion

This study investigated the presence, distribution and composition of microplastics (MP) particles in fishing grounds and fish species near the São Vicente Island of Cabo Verde. The findings revealed the widespread occurrence of MP in the marine environment, with fibers being the most commonly observed shape and black-colored particles being the most prevalent. The study suggests that inadequate waste management by individuals as well as fishing activities may contribute significantly to MP pollution in the area.

Analysis of fish samples indicated that the fish species did not exhibit selectivity regarding MP shape or color, ingesting whatever was available in their environment. This highlights the potential ecological implications of increasing MP abundance in marine waters. Despite the ingestion of MP by fish, no significant adverse effects on the growth condition of the fish were observed as a result of MP ingestion.

Polymer identification analysis revealed the presence of polyethylene (PE) and polypropylene (PP) as the dominant polymers in both fish and seawater, further implicating the possible sources such as textiles, packaging materials, as well as fishing gears as a significant source of MP pollution this environment.

The marketability of fish may face challenges if consumers become more and more aware of the potential dangers associated with consuming polluted fish products.

The potential threats that MP poses to the health of marketable fish species and their marketability calls for future research to gain a more comprehensive understanding of MP pollution levels and dynamics in fish caught in Cabo Verde.

7. Recommendation

This baseline study significantly contributes to the expanding body of knowledge on microplastics (MP) in marine environments, specifically in the Cabo Verdean marine environment. It highlights the critical importance of ongoing research, stakeholder collaboration, and the implementation of effective strategies to mitigate the adverse impacts of MP and protect the health and integrity of marine ecosystems.

To achieve a more comprehensive understanding of the sources, distribution, and ecological impacts of MP, future research should be conducted on a larger scale, utilizing larger sample sizes and longer monitoring periods. Additionally, further investigation should focus on the potential transfer of MP through the food web and assessing its long-term effects on marine organisms and the environment. Advanced techniques like Fourier Transform Infrared Spectroscopy (FTIR), capable of analyzing MP particles regardless of size, should be prioritized in future research endeavors.

Public awareness and educational programs on the risks and impacts of plastic pollution are crucial in promoting responsible consumer behavior, reducing plastic usage, and instilling environmental stewardship among citizens. Workshops, educational campaigns, and collaborations with local communities, including fishers, schools, and non-governmental organizations, are effective avenues for achieving this goal.

Furthermore, formulating and implementing policies addressing the use and management of plastics are of utmost importance. Such policies will play a significant role in combating the pervasive issue of plastic pollution and its detrimental effects on the ecosystem.

By integrating these recommendations, we can work towards mitigating the impacts of MP, preserving marine ecosystems, and ensuring a sustainable future for our oceans.

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Appendix

Table 3: The sampled fish species differed significantly in total length (cm). Results from ANOVA.

	sum_sq	df	F	PR(>F)
Fish species	852.8654	4.0	29.752706	4.832320e-16
Residual	680.7970	95.0	-	-

Table 4: Tukey's HSD test results for pairwise comparisons of fish total length (cm) among different species showing significant differences.

Group1	Group2	Mean diff	P-Adj	Lower	Upper	Reject
Caranx crysos	*Cephalopholis taeniops*	-0.305	0.9	-2.6591	2.0491	F
Caranx crysos	*Decapterus macarellus*	5.42	0.001	3.0659	7.7741	T
Caranx crysos	*Lithognathus mormyrus*	3.895	0.001	1.5409	6.2491	T
Caranx crysos	*Selar crumenopthalmus*	-2.53	0.0287	-4.8841	-0.1759	T
Cephalopholis taeniops	*Decapterus macarellus*	5.725	0.001	3.3709	8.0791	T
Cephalopholis taeniops	*Lithognathus mormyrus*	4.2	0.001	1.8459	6.5541	T
Cephalopholis taeniops	*Selar crumenopthalmus*	-2.225	0.0734	-4.5791	0.1291	F
Decapterus macarellus	*Lithognathus mormyrus*	-1.525	0.3799	-3.8791	0.8291	F
Decapterus macarellus	*Selar crumenopthalmus*	-7.95	0.001	-10.3041	-5.5959	T
Lithognathus mormyrus	*Selar crumenopthalmus*	-6.425	0.001	-8.7791	-4.0709	T

Table 5: The sampled fish species differed significantly in total weight (g). Results from ANOVA

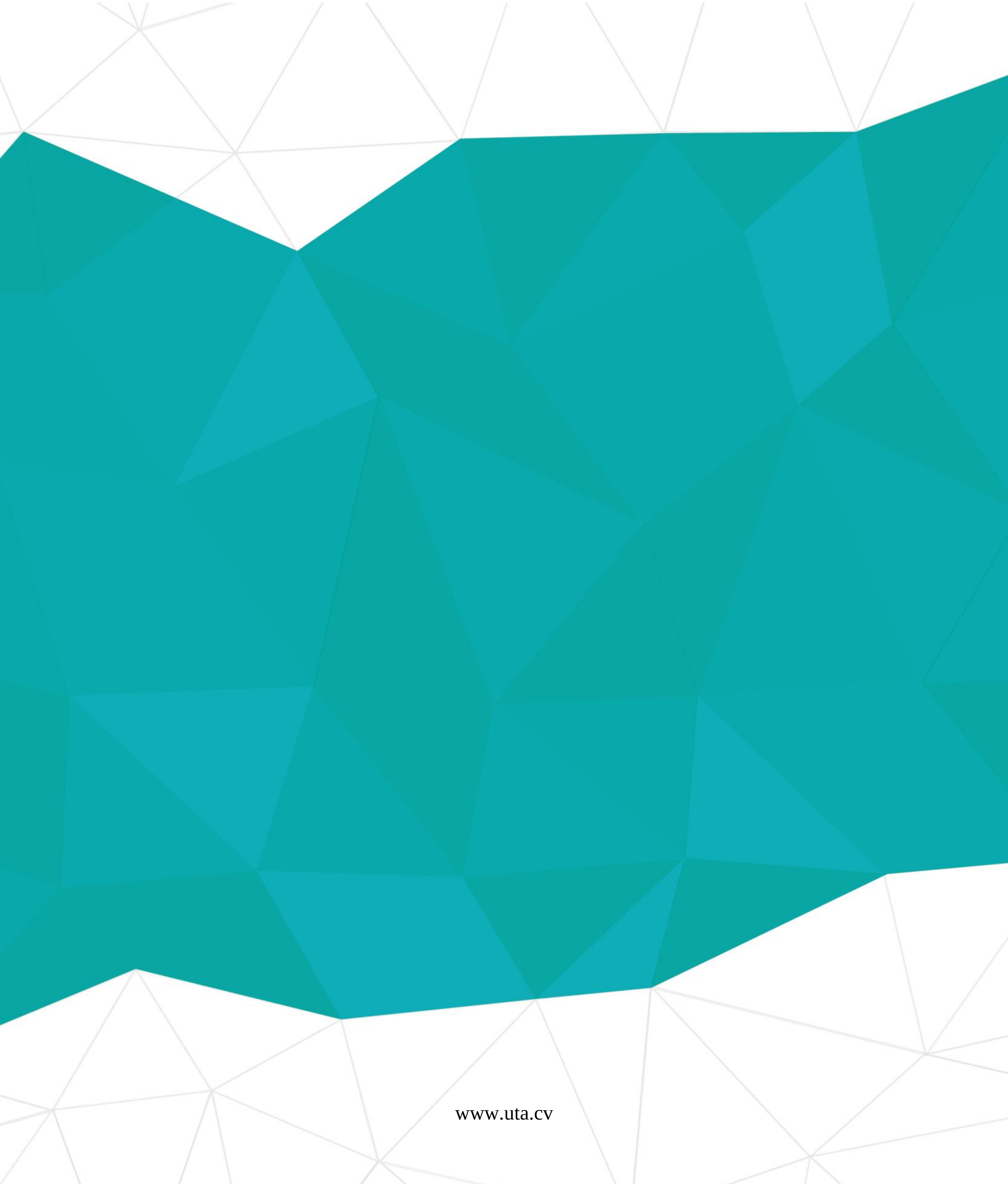
	sum_sq	df	F	PR(>F)
Fish species	678288.186210	4.0	21.035017	1.917619e-12
Residual	765834.612065	95.0	-	-

Table 6: Tukey's HSD test results for pairwise comparisons of fish total weight (g) among different species showing significant differences.

Group1	Group2	Mean diff	P-Adj	Lower	Upper	Reject
Caranx crysos	*Cephalopholis taeniops*	30.09	0.8037	-48.8669	109.0469	F
Caranx crysos	*Decapterus macarellus*	112.9545	0.0013	33.9976	191.9114	T
Caranx crysos	*Lithognathus mormyrus*	150.265	0.001	71.3081	229.2219	T
Caranx crysos	*Selar crumenopthalmus*	-81.5045	0.0396	-160.4614	-2.5476	T
Cephalopholis taeniops	*Decapterus macarellus*	82.8645	0.0348	3.9076	161.8214	T
Cephalopholis taeniops	*Lithognathus mormyrus*	120.175	0.001	41.2181	199.1319	T
Cephalopholis taeniops	*Selar crumenopthalmus*	-111.5945	0.0015	-190.5514	-32.6376	T
Decapterus macarellus	*Lithognathus mormyrus*	37.3105	0.661	-41.6464	116.2674	F
Decapterus macarellus	*Selar crumenopthalmus*	-194.459	0.001	-273.4159	-115.5021	T
Lithognathus mormyrus	*Selar crumenopthalmus*	-231.7695	0.001	-310.7264	-152.8126	T

Data availability

The data used in this study will be made accessible to the West African Science Service Center on Climate Change and Adapted Land Use (WASCAL) and will be available to all individuals who request access.



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