Occurrence of microplastics in commercial fish species from the Ethiopian rift valley's Lake Hawassa, Ethiopia

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Research Article

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Abstract

Microplastics (MPs) have recently been detected as emergent pollutants in the Ethiopian rift valley lakes located close to rapidly expanding towns. We provide the first study of MPs ingestion of commercial fish species from Lake Hawassa, Ethiopia: Catfish (*Clarias gariepinus*) and Nile Tilapia (*Oreochromis niloticus*). A total of 60 individual fish species was collected from three sampling sites of lake Hawassa in October 2020. Across all sampling sites, there was a significant difference in ingested MPs between benthic omnivore catfish and pelagic Phyto planktivorous Tilapia ($\chi^2 = 15.864$, $p < 0.001$). The most common size of ingested MPs (84.6%) was 0.5-1 mm, with fragments (59.5%) dominating, followed by fibers (25.4%). On average, 4.03 ± 1.33 MPs with sizes ranging between 60 μm and 10.53 mm were detected per individual. White and yellow MPs were particularly numerous, accounting for 36.8% and 26.4% of the total, respectively. Because Lake Hawassa's fishery is so significant, the potential impact of MP pollution on the lake biota in general, and economically valuable fish species in particular, deserves attention, additional research, and, if possible, early mitigation.

1. Introduction

Microplastics (MPs) are plastic particles that range in size from 1µm to 5mm in their longest dimension (SAPEA 2019). They are typically divided into two categories based on their origin, such as primary and secondary MPs (Pico et al. 2019). The principal MPs are purposefully created as plastic particles (Silva et al. 2018). They're also employed as abrasives in a variety of industries, including cosmetics, cleaning products, pharmaceuticals, and air-blasting media (Magnusson et al. 2016). Secondary MPs are the byproducts of the bigger plastic material breakdown by mechanical, microbiological, or photo-oxidative mechanisms (FAO 2019). They can also be produced during construction and as byproducts of plastics, such as road dust derived from tyre or road paint wear, or synthetic fibers shredded from fishing gear(Magnusson et al. 2016).

The presence of MP in water, sediment, and biota can have an impact on aquatic life. MPs, for example, reduce their ability to consume natural prey by obstructing the digestive tract (Cannon et al. 2016) or cause famine due to faux satiety (Cole et al. 2013), neurotoxicity, and oxidative damage (Barboza et al. 2018), and a decrease in photosynthetic activity and chlorophyll an of algae (Barboza et al. 2018; Y. Wu et al. 2019). MPs, on the other hand, can act as vectors for the transfer of xenobiotic chemicals and possible hazards to high trophic levels of animals via trophic transfer, putting food security and human health at risk (Carbery et al. 2018).

Urbanization and economic expansion are causing a massive increase in demand for plastics and plastic packaging, particularly in developing economies.However, there is little or no attention dedicated to managing plastic waste, particularly micro and macro plastics pollution awareness among the general public. As a result, fresh water and its biota are at risk of MP pollution.

Lake Hawassa, which is located directly adjacent to one of Ethiopia's fastest-growing cities, is significantly impacted by pollutants generated from industrial, municipal, and medical wastes. According to some research, high quantities of contaminants in industrial, municipal, and hospital wastes enter Lake Hawassa (Zinabu and Desta 2002; Bekele et al. 2021). Concerning microplastic pollution research, only one known case study, Lake Ziway, was explored on MP presence in fish and sediment (Merga et al. 2020), and (Jeevanandam et al. 2022) on MPS pollution in Lake Hawassa coastal sediment. However, evidence on the occurrence and extent of pollution of MPs in Ethiopian lakes, particularly Lake Hawassa, is still rare. To consider a solution to MPs contamination, it is necessary to be able to research their prevalence and abundance effectively. Thus, this is the first study to report the presence of microplastics in commercially and ecologically important fish species in Lake Hawassa, namely Tilapia (O. niloticus) and Catfish (C. gariepinus).

2. Materials And Methods

2.1 Description of the Study Area and site selection

2.1.1 Study area description

Lake Hawassa is located in the Sidama Regional State, near Hawassa city, approximately 275 kilometers south of Addis Abeba, at an elevation of 1680 meters in the middle Ethiopian rift valley (06°33'-07°33' N and 038°22'-038°29' E) (Fig. 1). The lake covers 90 km2 and has an average depth of 10.7 meters and a maximum depth of 22 meters (Tilahun and Ahlgren 2010). Six fish species live in Lake Hawassa, three of which are commercially important: Nile tilapia (*Oreochromis niloticus*, L, 1758), Catfish (*Clarias gariepinus*, B, 1822), and Big barb (*Labeobarbus intermedius*, R, 1836). (Dadebo 2000). Because of their small size, three more minnow species are not fished, according to this author: the straight fin barb (*Barbus paludinosis*), the black lampeye (*Aplocheilichthys* antinorii), and the stone lapping minnow (*Gara quadrimaculata*). Tilapia dominates the commercial fisheries, accounting for more than 85% of total yearly landings by weight, followed by catfish (14%), and large barb (1%) (Berehanu et al. 2015). The Tikur Wuha river, the sole perennial river that flows into Lake Hawassa, receives effluent from nearby factories/industries (placed on Hawassa's northern fringes) (Bekele et al. 2021). Municipal garbage and hospital wastewater enter the lake from the west, while shoreline agricultural waste enters the lake from the west and south.

2.1.2 Site Selection

The sampling sites are classified into three categories depending on the potential pollution sources (Fig. 1). Site 1, locally called (Tikur-wuha), which receives industrial effluent, is found on the lake's northwestern shore at $07^{0}06'$N Longitude and $038^{0}28'$E Latitude. Site 2, locally called "Amora Gedel" which receives municipal and hospital wastes, is found in the lake's central and southern sections, with coordinates of $07^{0}02'$N Longitude and $038^{0}27'$E Latitude. The third sampling site, locally known as

"Finch-wuha," receives Agrochemicals from shoreline cultivation, but is devoid of industry and city waste, found on the lake's southwest side, at $06^{0}59$'N Longitude and $038^{0}25$'E Latitude.

2.2 Sample selection

Two commercially important fish species (*O. niloticus* and *C. gariepinus*) were chosen for microplastic detection, The fact that the two species' niches and feeding areas differ provides some insight into the MPs' fate in the lake environment. As a result, studying these fish species has ramifications for economics, ecology, and possibly human health.

2.3 Sample Collection

A total of 60 individual fish were collected for analysis from fish landing places at the three sampling sites during the sampling period in October 2020. Fish caught by local fishermen who set their gill nets for tilapia and longlines for catfish at the selected sampling sites. The fish length that exceeded the lake's first maturity (C.gariepinus > 50cm, O.niloticus > 20cm) (Muluye et al. 2016) was chosen as the sample. For the analysis of gut content, adult individuals was used, so that the results could not be altered by possible variations in the diet of juvenile individuals (Abelha 2001). The fish samples were transported in an icebox to the department of biology laboratory of Hawassa College of Teacher Education and kept frozen until they were ready for analysis.

2.4 Sample extraction/digestion and separation

Body morphometrics was obtained after washing each fish specimen with distilled water. Following a longitudinal incision in the belly, the entire gastrointestinal tract (GIT) was dissected with ethanol-cleaned scissors and forceps from the buccal cavity to the anus. GIT was investigated in two techniques to extract possible plastic particles from the samples: 1) A visual examination of the entire gastrointestinal tract content was carried out utilizing a LEICA M250 C stereo microscope and LEICA DMC 4500 camera (Leica Camera AG, Switzerland). The first stage of sorting entailed a visual check of the entire gastrointestinal track, with anything that did not resemble natural prey being removed using forceps. and 2) digestion of the entire GIT content with 10% potassium hydroxide (KOH) solution in a volume ratio of 3:1 KOH to biological material, with samples incubated for 5 days (to allow full digestion of the GIT matrix at 60°C as indicated by (Bessa et al. 2018). According to (Zhao et al. 2014), using 10% KOH may decompose organic matter with an efficacy of more than 90%. The digested GIT was carefully sieved through a 0.1 mm sieve (the detection limit is 0.1 mm) (Merga et al. 2020), and the residue was transferred to a clean glass Petri dish for the second visual inspection using LEICA M250 C stereo microscope paired with a LEICA DMC 4500 camera to capture the picture and measure the length of MPS. Physical properties such as an artificial appearance, particle shape (e.g., Fragments, Fibers, Beads, Pellets, and Foam), and color were added as criteria (Vethaak and Leslie 2016). The malleability of the particles was assessed by squashing them with a stainless-steel dissecting needle (micro-tip diameter), as Cannon et al. (2016) performed to identify synthetic plastic from natural fibers.

2.5 Identification and polymer characterization of MPs

2.5.1 Visual identification

The filters were examined with a Leica M 250 C stereo microscope, and pictures, microplastics count, and MP size were acquired with a Leica DMC 4500 digital microscope camera. The sizes were established by measuring the length of their longest axis, as reported by Li et al. (2018) dividing them into four classes: 0.5mm, 0.5-1mm, 1-2.5mm, and 2.5-5mm. According to Li et al. (2018), and Lusher et al. (2017), microplastics are divided into five types: fragments, fibers, beads, pellets, and foam.

2.5.2 Characterization of MPs

The polymer type was determined using a Spectrum 65 FT-IR (PerkinElmer) at the Addis Ababa University chemistry laboratory. The measurement resolution was set at 4 cm-1 in the 4000 – 400 cm-1 range, with four scans using KBr windows. Identification with the reference library was assigned for matches larger than 75% (Reinold et al. 2021).

2.6 Data quality assurance and quality control

A series of measurements were made in the laboratory to avoid background contamination. The experiment was carried out while wearing cotton lab coats and gloves. All of the solutions in the study were passed through a 0.45µm filter before being employed in the trials. Tools, glassware, and containers were carefully cleaned with deionized water and wrapped in aluminum foil after each stage. To avoid additional contamination, all synthetic clothes was refused throughout the microscopic investigation.

2.7 Data Analysis

The Shapiro-Wilk test was done to check that the data was normal. SPSS 26 was used to analyze the data, and Origin Pro 8.5 was utilized to plot the spectra peak of sample polymers. The data were evaluated using descriptive statistics such as frequency distribution, percentage, mean, and standard deviation to determine the occurrence of microplastics in the GIT. Chi-squared tests were used to examine the relationship between dependent and independent variables. Pearson and Spearman rho correlation analysis was used to evaluate potential relationships between the presence of microplastics and the overall length and weight of fish in the sampling site. Statistical tests were deemed significant at P 0.01 and 0.05.

3. Results And Discussion

3.1 Habitat and morphometrics of sampled fish species

Table 1
Habitat, Length and Weight of sampled fish

Fish species	Habitat	Feeding habit	Sample size (n)	Average Length ±SD (cm)	Average weight ± SD (g)
C. gariepinus	Benthic	Omnivorous	30	55.77 ± 3.81	902.15 ± 12
O. niloticus	Pelagic	Phytoplanktivorous	30	30.20 ± 0.91	362.30 ± 29

Table 1 shows habitat and morphometric data from three sample sites for Catfish (*Clarias gariepinus*) and Nile Tilapia (*Oreochromis niloticus*). The Catfish, which is benthic and feeds omnivorously, measured an average of 55.8 ± 3.8 cm and varied in length from 50 to 60 cm. Catfish weighed 982.2 ± 12g on average and ranged from 680g to 1.02kg. Nile Tilapia is a pelagic and phytoplanktovorous fish that ranges in size from 28.5cm to 32cm. Nile Tilapia weighed between 320g and 420g on average. In general, both fish species employed for MP detection in their GIT were adults based on their length and weight.

3.2 Occurrence and abundance of MPs

3.2.1 Ingestion of MPs by sampled fish

Thirty Nile Tilapia and thirty catfish were taken from Lake Hawassa to test the presence of MP in their GI tracts. According to Table 2, MPs were found in the GIT of 37 of the 60 fish tested (61.7%). This proportion is equal to the 66% of freshwater salmonids observed by O'Connor et al.(2020) in the Irish riverine system, the 68% of the Swedish West Coast by Karlsson et al.(2017), and the 69.7% of fishes obtained from a city water supply in Lake, Southwestern Nigeria by Adeogun et al.(2020). MPs were found in 11 (36.7%) and 26 (86.6%) tilapia and catfish samples, respectively, as reported in Table 2. This finding was comparable with the reported 35% for tilapia taken from Lake Victoria (Biginagwa et al. 2016). However, the current finding was lower than the report of Merga et al. (2020) for tilapia (22%) and catfish (41%), both from Lake Ziway. The discrepancy in results could be due to different sources of MPs in the lake as well as a difference in sample settings. The pooled data on individual fish per site (Table 2) revealed that the number of Catfish that ingested MPs was significantly more than the number of Tilapia in all sites ($\chi 2 = 15$, $p < 0.001$). This is attributed to their habitat and feeding habit. Catfish are benthic omnivorous while Tilapia is pelagic phytoplanktivorus. A similar result was reported by other investigators (Bour et al. 2018; Merga et al. 2020).

Table 2
Number of Fish species ingesting MPS per site(n = 30)

Targeted fish species	Sample size (n)	Total number of fish with MPS(%)	Number of fish with MPs per site		
			Site 1	Site 2	Site 3
C.gariepinus	30	26	10	9	7
O. niloticus	30	11	4	3	4
Total	60		14	12	11

3.2.2 Abundance of MPs in GIT of sampled fish

Table 3 shows that a total of 402 plastic particles were counted from the GIT of 37 fish sampled from three sites, with an average abundance of 4.03 ± 1.33 plastics per individual fish per site. The average number of MPs consumed did not differ significantly among species or sample sites (χ^2, $P > 0.05$). This demonstrates the widespread prevalence of microplastic contamination in Lake Hawassa, which is linked to industrial, municipal, and agricultural wastes disposed of near the sampling sites. In addition, Site 2 (Amora Gedel Site) had the highest total MPs for both species, with 113 MPs for catfish and 91 MPs for tilapia, showing that the site received a significant number of MPs from municipal and urban wastes. This result is in line with the Stockholm International Water Institute (SIWI), (2020) research, which said that 68% of all plastic litter generated in urban and tourist areas are not collected for recycling, which might be a source of MPS on the site. Overall, the current study's findings are consistent with those of (Horton et al. 2018; Lusher et al. 2017), who discovered that microplastics are more numerous in freshwater bodies surrounding urban and industrial centers, as well as irrigated farmlands.

Table 3
Abundance of MPs (Mean + SD) count/individual/site detected in the GIT of sampled fish species

Fish species	Total	Fish with ingested MPs	Site	Average count/ individual/site	Max.count/ individ/site	Total count
C.gariepinus	30	26	1	2.85 ± 1.69	6	74
			2	4.38 ± 3.25	34	113
			3	4.86 ± 3.11	10	68
O. niloticus	30	11	1	2.57 ± 1.40	4	18
			2	6.07 ± 5.08	15	91
			3	3.45 ± 3.05	8	38
Total	60	37	**Average**	4.03 ± 1.33		402

According to Table 4, there was a statistically significant positive and moderate association between the abundance of MPs ingested by fish TL (Spearman's rank correlation, rho = 0.525; $p < 0.001$), TW (Spearman's rank correlation, rho = 0.487; $p < 0.001$), and for both habitat and feeding habit (Spearman's rank correlation, rho = 0.514; $p < 0.001$). This implies that as the fish size or weight increases the potential to ingest MPs particles also increases. This finding was inline with other researchers, including Park et al. (2020) from Korea, (Horton et al. 2018; McNeish et al. 2018) from the United States.However, some research have found no link between microplastic concentration

Table 4
Correlations of MPs occurrence with fish length (cm), body weight (gm), habitat, feeding mode, species, and sample site

			Occ	TL	Weight	Habitat	F.habit	Species	Site
Spearman's rho	MPs Occ	Corr. coeff *(r)*	1.00	**.525***	**.487***	**.514***	**.514***	**.514***	-0.42
		Sig. (2-tailed)		**0.000**	**0.000**	**0.000**	**0.000**	**0.000**	0.**750**

occurrence and fish length or weight (de Vries et al. 2020).

3.3 Physical characteristics of MPs detected

According to the report of Aragaw (2021),MPs particles have no distinguishing characteristics; rather, they have a variety of physical, chemical, and structural properties. Accordingly, in the present study MPs were classified based on their physical characteristics (shape,color and size) identified through visual observation under a stereomicroscope equipped with a camera as follows.

3.3.1 Shapes of MPS detected

In the current investigation, MPs recovered from the GIT of the studied fish were classified into the following shapes: fragments, fibers, beads, pellets, and foam (Fig. 2). Identifying the morphologies of MPs' characteristics is crucial for the future establishment of plastic waste management, because the MPs found are a direct sign of virgin macro-plastics.

As shown in Fig. 3, the most commonly observed microplastics are fragments, which account for 59.5% of all microplastics, followed by fibers and beads, which account for 25.4% and 8.2%, respectively, and other types account for 6.9% of all MPs. Plastic bags and containers abandoned fishing nets, and food wrappers were often observed during field sampling at both sample sites. This observation was confirmed by the Stockholm International Water Institute (SIWI) (2020) research, which says that fishing nets in Lake Hawssa are frequently disposed of directly into the lake as a convenient disposal method and also to conceal them from authorities. This illegal activity may lead to the fragment's dominance in the lake. The finding of fragment dominancy was consistent with the findings of Boerger et al. (2010) from the North Pacific Central Gyre types of polymers retrieved from fish GIT, Tanaka and Takada (2016) from Japan recorded plastic debris in Japanese anchovy, Merga et al. (2020) from Ethiopia of Lake Ziway freshwater fishes, and Eriksen et al. (2013) from the upper mid-east region of North America surface saltwater.

In terms of spatial variability in the types of MPs ingested by fish, the current study discovered that the Tikuer-wuha site (Site 1 with 13.9% fiber type) and Finch-wuha (Site 3 with 25% fragment) had a significantly (**χ^2 =** 31.876; P < 0.001) higher frequency of fish ingested MP particles than the other two sites (see **SI.1 Fig**). The fragment type could be related to the fragmentation of white plastic materials used to support tomato plants. This result is consistent with the findings of Merga et al. (2020) from Lake Ziway, who claim that the fragment type of white plastic items is associated with tomato cultivation. The second most prevalent MP is fiber type, as earlier research indicates that the fabric and textile industry may contribute fiber to water bodies and aquatic species (Katare et al. 2021). Furthermore, several factories in the current study site have wastewater treatment plants (WWTPs), but research shows that no wastewater treatment technique completely removes microplastics, hence WWTPs are regarded as point sources of MPs release into the aquatic environment (Habib et al. 2020).

3.3.2 Color of MPs detected

As shown in Fig. 4, the most common color of MPs identified in the GIT of sampled fish was white (36.8%), followed by yellow (26.4%), and transparent (14.7%). The remaining seven hues accounted for less than 22% of the MPs found in the research fish's GIT. The current result of high white color was compatible with Merga et al (2020) and Sun et al. (2021). Yellow was the second most prominent color in this study, despite the fact that Reinold et al (2021) identified yellow as the dominant color of MPs. The predominance of white-colored MPs may be related to the types and quantities of plastics (MPs) utilized locally (Zhou et al. 2020). Furthermore, on-site inspections revealed that coastal tomato production was prevalent, and growers used white plastic objects to support these plants; hence, the current white color dominance of MPs may be ascribed to such human actions. Similarly, the existence of degraded plastic items could be linked to the second dominance of yellow MPs, since Liu et al. (2019) discovered that yellow MPs are the outcome of degraded products.

Colors of ingested microplastic particles varied significantly across three sampling sites (Pearson's correlation (r) = -0.129; p = 0.009) and species (Pearson's correlation (r) = -0.109; p = 0.029), Site 1 (Tikuer-wuha) had a disproportionately high quantity of colored microplastics (white 16.4% and yellow 20.2%) when compared to the other two locations. This is most likely due to industrial effluent discharged into the Tikur Wuha River by industries like textile, ceramics, soft drink, and beer factories on Hawassa's eastern fringes.

3.3.3. Size of MPs detected

The MP's size distribution ranged from 60μm to 10.53mm. The most common size was more than 0.5-1 mm (85% of the total number of MPs) (**SI.2Fig**). The largest MPs was fiber type. There was no statistically significant difference between sampling sites and species (Pearson's correlation (r) = 0.05; p = 0.301). This demonstrates that microplastics from all sources affected both fish species. Our findings were consistent with a recent study of microplastic pollution along Lake Hawassa's shores (Jeevanandam et al. 2022).

3.4 Polymer composition of identified MPs

Polystyrene (PS), Polypropylene (PP), Low-density polyethylene (LDPE), High-density polyethylene (HDPE), Polyamides (all Nylon), Polycarbonate (PC), Poly-methyl methacrylate (PMMA or Acrylic), Ethylene-vinyl acetate (EVA), and Acrylonitrile butadiene styrene were detected in the current study (**SI.3Table**). This result was consistent with the Stockholm International Water Institute's (SIWI) (2020) investigation. Based on the investigation report, the types of plastic generated in the Lake Hawassa basin include dense/heavy (PET, LDPE, HDPE, PP), film/light (bags and wrappers), and fishing nets. Furthermore, the results of PP and PE are consistent with those of Corcoran et al. (2015), who researched Lake Ontario in North America. The identification of these polymers allows for speculation on the origins of various polymers in the studied area. Several authors (Lusher et al. 2017; Wu and Munthali 2021) have suggested that the identified polymers PE (HDPE and LDPE), PS, and polyamide (PA) in the study area may have originated from fishing activities or fishing equipment, such as nets, traps, hooks, lines, plastic bags, and bottles thrown into the area around freshwater bodies of water. Other polymers, such as PET, PVC, Acrylic or Rayon, and CA/cellophane, are used in the textile sector to make non-woven fabrics, apparel, and carpets, as well as milk jugs, plastic bottles, packing bags, drinking straws, and bottles for beverages other than water (Renzi and Blašković, 2018;Reinold et al. 2021). Thus, the aforementioned polymer sources may be suspected in urban lakes such as Lake Hawassa, where agricultural pollutants and industrial effluents may enter the lake through a number of entrance channels such as wastewater drainages (municipal wastes). Furthermore, recreational activities such as boating, vacationing, and fishing may be substantial contributors to the number of microplastics, as seen by Merga et al. (2020) in Lake Ziway

4. Conclusion

The current work is the first to detect and assess the presence of microplastics in the GIT of two economically important fish species from Lake Hawassa, *O. niloticus* and *C. gariepinus*. According to the findings, both fish species are vulnerable to microplastics contamination. However, because of their habitat and feeding habits, benthic and omnivorous Catfish (*C. gariepinus*) are more vulnerable to MP pollution. As a result, the presence of MPs in commercially and ecologically important fish species inhabiting Lake Hawassa, as well as their widespread abundance in different sampling sites, calls for early intervention and better waste management, with a focus on major routes of pollutants entering the lake.

Declarations

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could appear to have influenced the work described in this paper.

CRediT: authorship contribution statement

Asrat   Fekadu: Conceptualization; Methodology; Investigation, Formal analysis Project and investigation; Writing - original draft preparation; Writing - review and editing

Girma Tilahun: Supervision; Formal analysis and investigation; Writing - review & editing.

Availability of data and materials

The data sets on which the conclusions of the paper rely is presented in the main body of the manuscript.

Competing interests

The authors declare that they have no competing interests.

Consent for publication

Not applicable.

Ethics approval and consent to participate

Not applicable.

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Supplimantary Information(SI)

SI 1 Figure. Shapes of Micro plastics per site

SI 2 Figure. Size of MPS detected

SI 3 Table. The site, species, and FTIR characteristic

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Figures

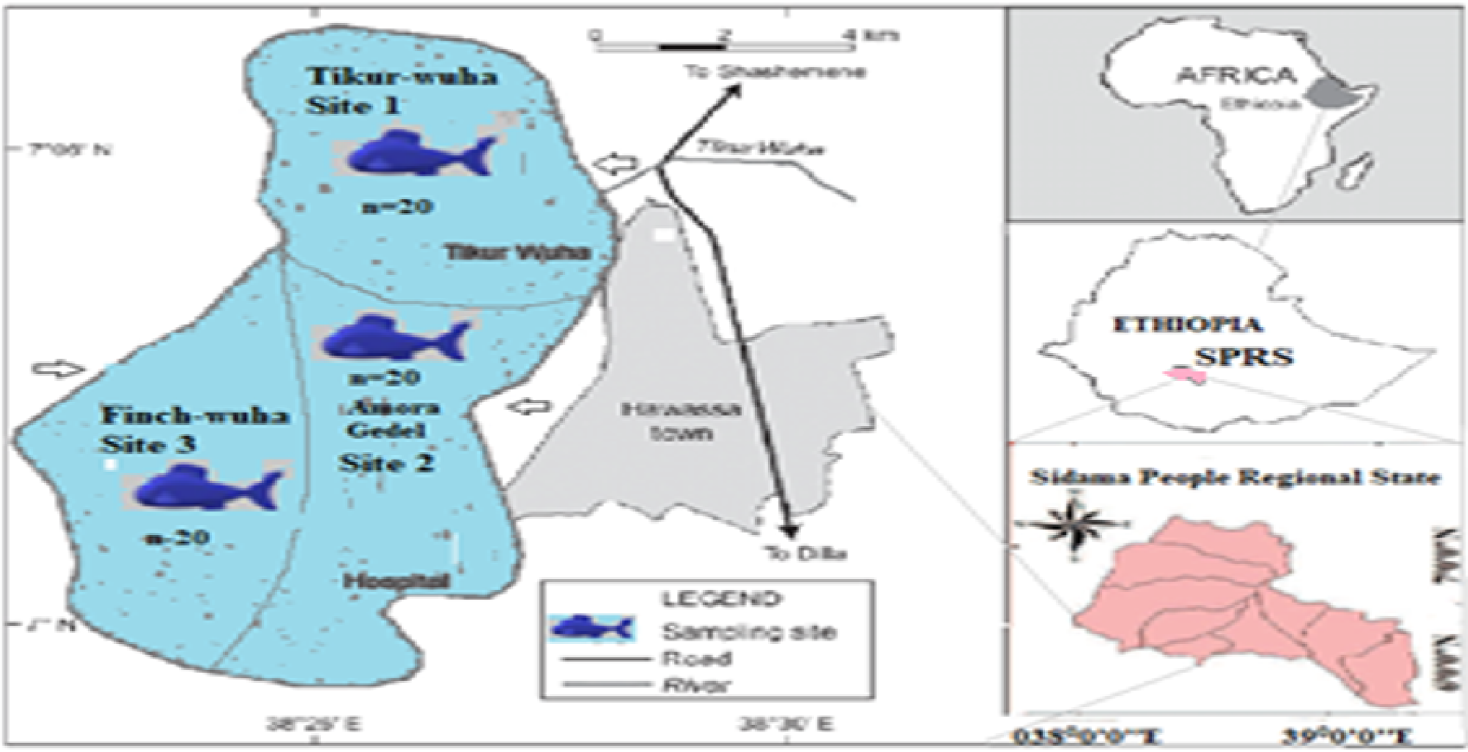

Figure 1

Map of Lake Hawassa modified after Sorsa et al.(2016) and Yaekob Chiriko (2021)

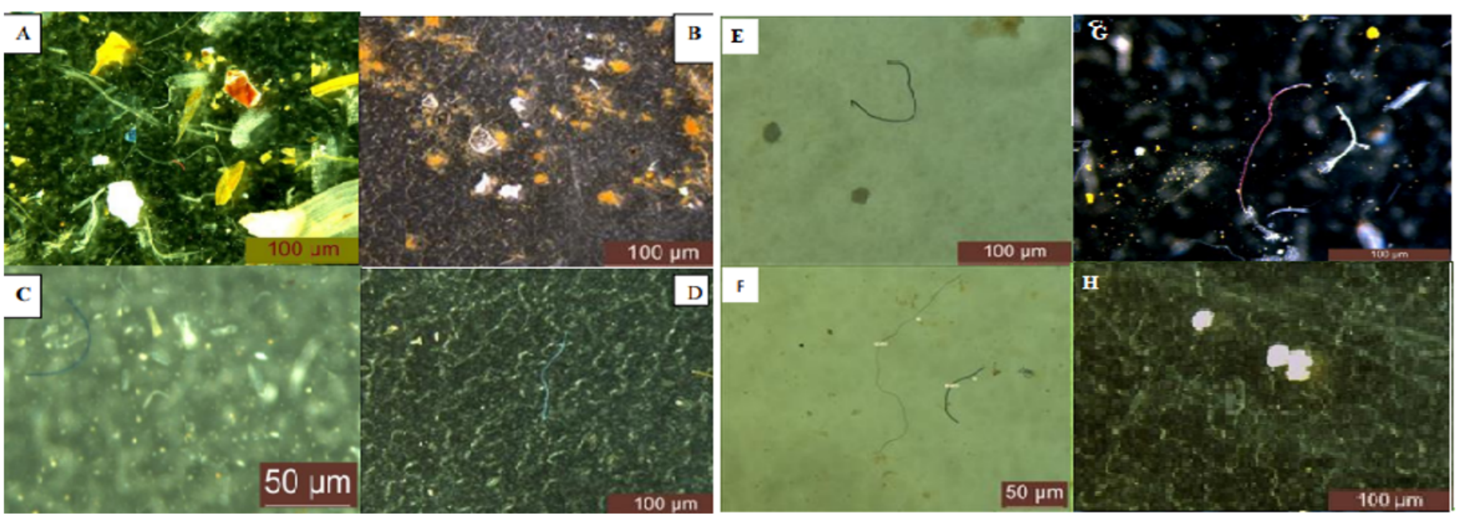

Figure 2

Types of MPs detected in the GIT of Nile-Tilapia and Catfish: Fragment (A,B),fiber(D-G),and Pellets(H).

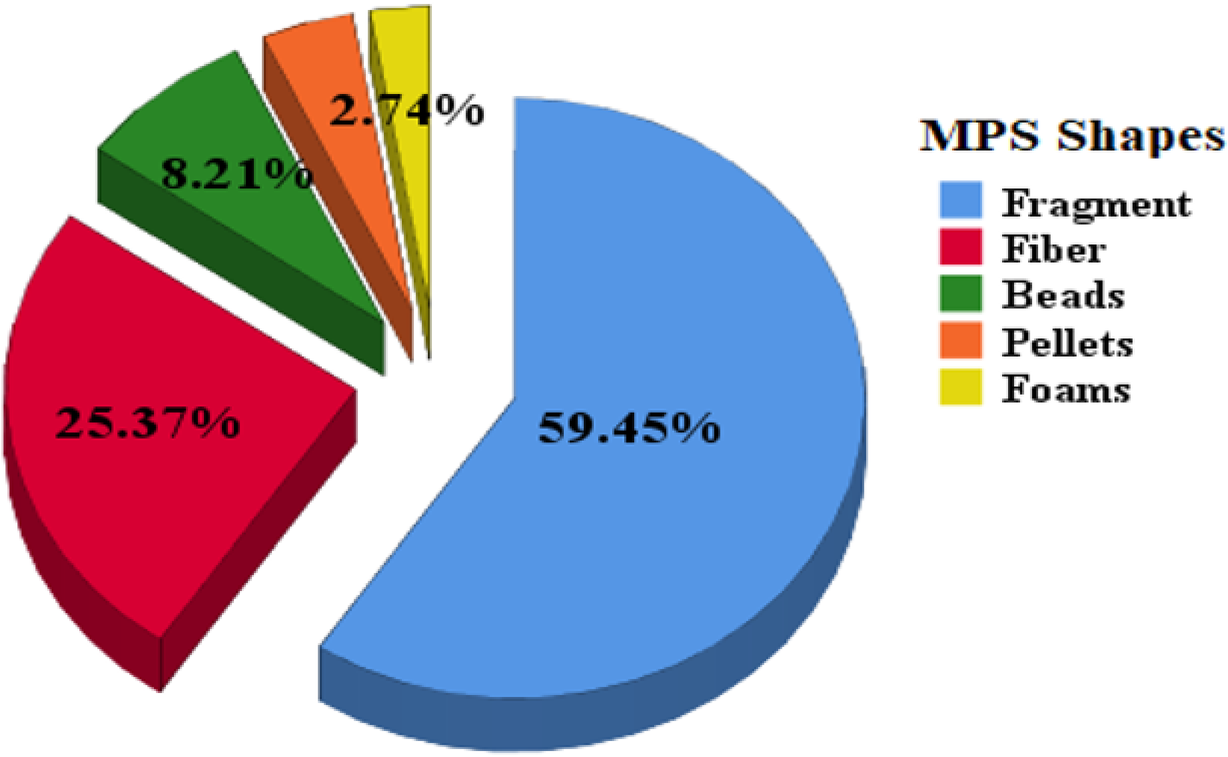

Figure 3

Shape of MPS of the fish samle

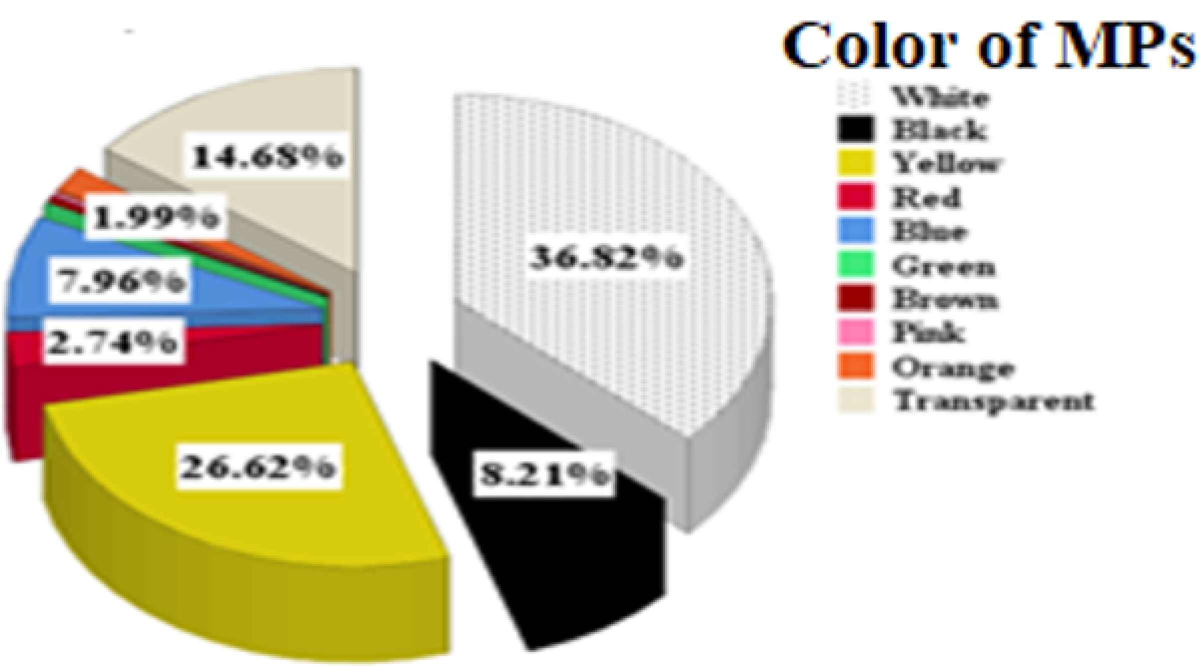

Figure 4

Color of the MPs in the fish sample

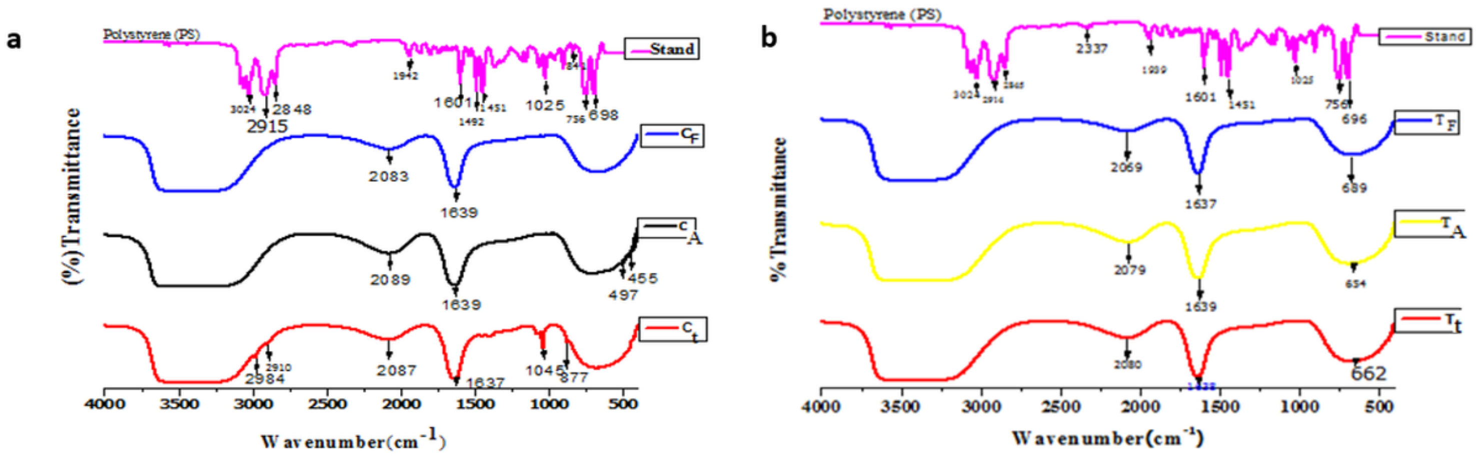

Figure 5

(a and b). FTIR plot spectra peak of Catfish, and Tilapia sample MP polymers, respectively and the reference spectra of Polystrene (PS).

Key:C=Catfish,F=Finch-wuha,A=Amora-Gudel,t=Tikuer-wuha;T=Tilapia

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