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Assessment of Microplastic Contamination and its Ecological Impact on the Aquatic Ecosystem of River Sone at Old Shahabad

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Abstract: Microplastic pollution has emerged as a pervasive environmental threat in freshwater ecosystems, yet data from Indian rivers remain limited. This study investigates the ingestion and trophic transfer of microplastics across plankton, benthic macroinvertebrates, and fish in the River Sone, Bihar, India, focusing on midstream (Dehri-on-Sone) and downstream (Koilwar) sites. Biological samples were collected seasonally (pre-monsoon, monsoon, post-monsoon) and analyzed for microplastic abundance, morphology, size, and polymer composition using stereo microscopy, FTIR, and Raman spectroscopy. Microplastics were detected in all trophic levels, with the lowest abundance in plankton (2.1–6.4 particles m^{-3}) and the highest in fish (7.2–28.9 particles $individual^{-1}$), indicating trophic transfer and accumulation. Fibers were the dominant morphology, and polyethylene, polypropylene, and polystyrene were the most common polymers. Seasonal variation revealed elevated microplastic ingestion during pre- and post-monsoon periods, reflecting hydrological influences on particle availability and sediment resuspension. The results highlight the role of benthic macroinvertebrates as intermediate reservoirs, facilitating microplastic transfer to higher trophic levels. Continuous ingestion poses potential sub-lethal physiological stress, ecological disruption, and human health risks, especially considering the consumption of local fish. This study provides the first evidence of trophic amplification of microplastics in the River Sone and underscores the urgent need for continuous monitoring, improved waste management, and policy interventions to mitigate plastic contamination in Indian freshwater ecosystems. The findings contribute to a broader understanding of microplastic dynamics in tropical riverine environments and their implications for freshwater biodiversity and food security.

Key points: Microplastics, Trophic transfer, Freshwater ecosystems, River Sone, Polymer composition, Seasonal variation.

1. Introduction

Microplastics, commonly defined as plastic particles smaller than 5 mm in diameter, have emerged as one of the most pervasive and persistent contaminants in aquatic environments worldwide (Thompson et al., 2004). These particles originate either as primary microplastics, intentionally manufactured at small sizes for industrial and consumer applications, or as secondary microplastics formed through the fragmentation of larger plastic debris under physical, chemical, and biological processes (Andrady, 2011). Due to their small size, low density, and resistance to degradation, microplastics are easily transported through aquatic systems and can persist for extended periods, posing long-term ecological risks.

While microplastic pollution has been extensively studied in marine ecosystems, freshwater environments particularly rivers have received comparatively less scientific attention (Wagner et al., 2014). Rivers play a dual role as both sinks and conduits for microplastics, transporting large quantities of plastic debris from terrestrial sources to downstream lakes, reservoirs, and ultimately oceans (Lebreton et al., 2017). Urbanization, improper waste management, wastewater discharge, agricultural runoff, and industrial effluents significantly contribute to microplastic loading in riverine systems, especially in developing countries where regulatory enforcement is often limited (Blettler et al., 2018). Freshwater biota are particularly vulnerable to microplastic exposure due to continuous contact with contaminated water and sediments. Microplastics have been documented in a wide range of freshwater organisms, including plankton, benthic macroinvertebrates, and fish (Li et al., 2020). Lower trophic organisms such as phytoplankton and zooplankton can ingest microplastics directly because of their small size and non-selective feeding mechanisms (Cole et al., 2013). Once ingested, microplastics may cause physical blockage, reduced feeding efficiency, oxidative stress, and altered growth and reproduction in planktonic communities, potentially disrupting primary productivity and nutrient cycling (Wright et al., 2013).

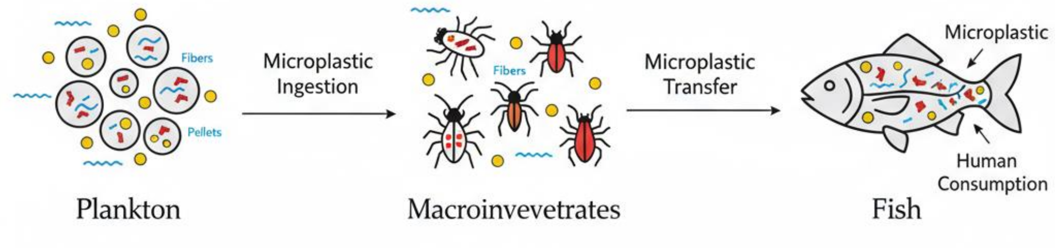

Fig.1: Conceptual Diagram of Tropic Transfer of Microplastics

Benthic macroinvertebrates represent a crucial link between primary producers and higher trophic levels in freshwater food webs. These organisms often inhabit sediments where microplastics tend to accumulate, making them highly susceptible to ingestion through deposit-feeding or detritivorous behavior (Hurley et al., 2018). Studies have demonstrated that macroinvertebrates can retain microplastics for prolonged periods, increasing the likelihood of transfer to predators such as fish (Imhof et al., 2017). Such trophic transfer is of particular concern because it facilitates the movement of microplastics and associated toxic additives or adsorbed pollutants across trophic levels. Fish, as higher trophic consumers, are considered effective bioindicators of microplastic pollution in freshwater ecosystems. Numerous studies have reported microplastic ingestion in freshwater fish species, with particles detected in gastrointestinal tracts and, in some cases, translocated to other tissues (Roch et al., 2020). Microplastic ingestion in fish has been associated with inflammatory responses, metabolic disturbances, and altered behavior, raising concerns regarding ecosystem health and food safety (Lu et al., 2016). The potential for microplastics to act as vectors for heavy metals, pesticides, and pathogenic microorganisms further amplifies their ecological and toxicological significance (Rochman et al., 2013).

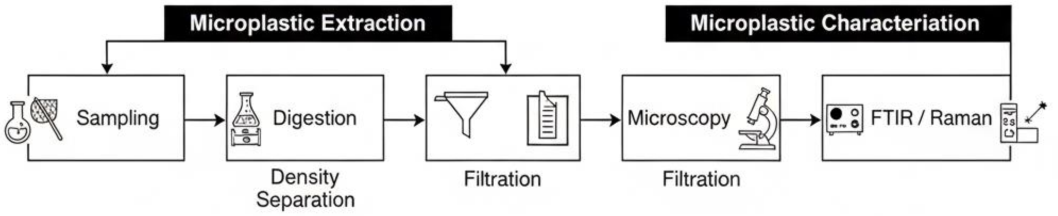

Fig. 2: Schematic workflow of microplastic extraction and characterization from freshwater biota

Despite growing global awareness, data on microplastic contamination in Indian freshwater systems remain scarce. Most existing studies in India have focused on coastal and marine environments, while large river systems such as the Ganga and its tributaries are still underrepresented in scientific literature (Kumar et al., 2021). The River Sone, a major southern tributary of the Ganga, flows through ecologically and socioeconomically important regions of Bihar. The river supports diverse aquatic biodiversity and provides essential ecosystem services, including fisheries, irrigation, and domestic water supply. The increasing population pressure, urban discharge, plastic waste mismanagement, and agricultural activities have raised concerns about emerging pollutants, including microplastics, in the River Sone ecosystem. The present study aims to assess the ingestion, distribution, and trophic transfer of microplastics across plankton, benthic macroinvertebrates, and fish in the River Sone ecosystem. By analyzing gut contents and characterizing microplastic types, this research seeks to provide baseline data on freshwater microplastic contamination in eastern India and contribute to the broader understanding of microplastic dynamics in riverine food webs.

2. Literature Review

Microplastics plastic particles smaller than 5 mm have gained global attention as pervasive pollutants in aquatic ecosystems due to their durability, ubiquity, and potential to disrupt ecological processes and food webs (Bhardwaj et al., 2024). Historically, research on microplastics focused primarily on marine systems, driven by their visibility and ease of sampling in coastal waters (Thompson et al., 2004; Meijer et al., 2021). The scientific community has increasingly recognized the importance of freshwater environments, particularly rivers, as both sinks and conduits for microplastic transport (Neelavannan & Sen, 2023; Kumar et al., 2025). Freshwater ecosystems differ from marine systems in several important ways that influence microplastic dynamics. Due to their proximity to terrestrial sources such as urban waste, industrial effluents, agricultural runoff, and sewage discharge rivers often exhibit higher concentrations of microplastics that vary spatially with human population density and land-use patterns (India Water Portal, 2025). These particles are typically present as primary microplastics manufactured at microscopic sizes, or more commonly as secondary microplastics formed through the fragmentation of larger plastic debris (Neelavannan & Sen, 2023).

A key concern in freshwater research is how microplastics interact with biological communities, especially across trophic levels. Evidence suggests that microplastics can be ingested by multiple taxonomic groups, initiating at the lowest trophic levels such as plankton and progressing to macroinvertebrates and fish (Wagner et al., 2014; Eerkes-Medrano et al., 2015). Microplastics readily adhere to or are ingested by phytoplankton and zooplankton due to their small size and non-selective feeding mechanisms, potentially impairing feeding efficiency, growth, and reproductive success (Cole et al., 2013; Eerkes-Medrano et al., 2015). The ingestion of microplastics by lower trophic organisms facilitates their entry into the food web. Planktonic consumption can lead to microplastics being transferred to benthic macroinvertebrates through predation and detrital pathways (Hurley et al., 2018). Macroinvertebrates, being closely associated with sediments where microplastics often accumulate, are particularly susceptible to exposure. These communities act as an essential link between primary producers and higher trophic consumers such as fish, amplifying the potential for trophic transfer.

The concept of trophic transfer refers to the sequential movement of contaminants from one trophic level to the next, and evidence is mounting that microplastics can follow such pathways in freshwater systems. A recent systematic review highlighted that microplastic ingestion has been confirmed across multiple freshwater taxa, albeit the frequency of studies demonstrating actual trophic transfer remains low (only about 12% of publications reviewed) (Reynolds et al., 2021). The scarcity of multi-level trophic studies underscores a significant knowledge gap, particularly concerning long-term bioaccumulation patterns and ecological implications in freshwater food webs. Fish, as apex consumers in many freshwater ecosystems, have been extensively studied for microplastic ingestion. Field studies demonstrate widespread microplastic ingestion in fish, with particles often detected in gastrointestinal tracts and sometimes even in other tissues (Roch et al., 2020). Microplastics can cause adverse physiological effects, such as inflammation, oxidative stress, and energy depletion, which may compromise growth and survival (Lu et al., 2016).

Microplastics often adsorb persistent organic pollutants (POPs) and heavy metals, thus acting as vectors for additional toxicants that can exacerbate their impact on aquatic biota (Rochman et al., 2013).

Despite global interest, research on freshwater microplastics in India remains comparatively sparse. Much of the existing literature focuses on microplastic contamination in sediments, surface water, and biota of major rivers like the Ganga, Yamuna, and Brahmaputra (India Water Portal, 2025; Neelavannan & Sen, 2023). For example, Kolavai Lake in Tamil Nadu exhibited significant interactions between microplastics and zooplankton, revealing negative correlations suggestive of ecological disruption (Ajay et al., 2023). Similarly, research on indigenous freshwater fishes in India documented microplastic contamination across multiple fish species, highlighting potential risks to food safety and ecosystem health (Singh et al., 2022).

The River Sone have received limited scientific scrutiny, particularly concerning multi-trophic microplastic ingestion and transfer. Given the river’s socio-economic importance for irrigation, fisheries, and domestic water supplies, understanding microplastic dynamics in its biotic communities is essential for effective ecosystem management and pollution mitigation strategies. Comprehensive investigations that link microplastic occurrence with trophic transfer are urgently needed to inform policy and conservation efforts in Indian freshwater systems.

3. Materials and Methods

2.1 Study Area

The present investigation was carried out along the Bihar stretch of the River Sone, a major southern tributary of the River Ganga, which plays a crucial role in regional hydrology, fisheries, irrigation, and domestic water supply. Two sampling locations were selected to represent contrasting hydrological and anthropogenic conditions: Dehri-on-Sone (midstream) and Koilwar (downstream).

Dehri-on-Sone (24.90°N, 84.18°E) represents a midstream segment characterized by moderate urbanization, municipal wastewater discharge, small-scale industrial activities, and transportation-related plastic inputs. Koilwar (25.60°N, 84.80°E), located downstream, receives cumulative pollutant loads transported from upstream regions and is influenced by intensive agricultural practices, fishing activities, and seasonal sediment deposition. The downstream stretch is particularly prone to microplastic accumulation due to reduced flow velocity and sediment trapping during post-monsoon periods. Sampling was conducted during pre-monsoon (March–May), monsoon (July–September), and post-monsoon (October–December) seasons to account for hydrological variability. Seasonal discharge variation (Q) influences microplastic transport according to the relationship:

$$MP_{flux} \propto Q \times C_{MP}$$

where MP_{flux} is microplastic flux (items day-1) and C_{MP} is microplastic concentration (items m^{-3}), as reported in riverine transport studies (Lebreton et al., 2017; Wagner et al., 2014).

2.2 Sample Collection

2.2.1 Plankton Sampling

Plankton samples were collected using plankton nets of 20μ m and 60μ m mesh sizes, allowing the capture of both phytoplankton and zooplankton communities. Horizontal surface tows were conducted for $5-10$ minutes against the river current. The volume of filtered water (V) was estimated using:

$$V = A \times d$$

where A is the mouth area of the net (m^2) and d is the distance covered during towing (m). This enabled the calculation of microplastic concentration as:

$$C_{MP} = \frac{N_{MP}}{V}$$

where N_{MP} represents the number of microplastic particles counted.

Collected plankton samples were transferred to pre-cleaned glass bottles using filtered river water and preserved in 4% formalin. Plastic materials were strictly avoided during sampling to prevent contamination. Prior to microplastic extraction, plankton abundance and taxonomic composition were documented to examine potential relationships between feeding strategy and microplastic ingestion (Cole et al., 2013; Li et al., 2020).

2.2.2 Benthic Macroinvertebrate Sampling

Benthic macroinvertebrates were collected using kick nets (500 µm mesh size) following standardized freshwater biomonitoring protocols. The sampling involved disturbing the substrate upstream of the net for a fixed duration (2–3 minutes), allowing organisms to be dislodged into the net. Specimens were obtained by handpicking from submerged stones, aquatic vegetation, and sediment surfaces to ensure representative coverage of functional feeding groups.

Table 1. Sampling methods and sample size across trophic levels

Biotic Group	Method	Mesh/Tool	Sample Size (n)
Plankton	Net tow	20 & 60 µm	18
Macroinvertebrates	Kick net	500 µm	24
Fish	Gill/Cast net	—	30

Specimens were sorted in the field using stainless steel forceps and preserved in 70% ethanol. In the laboratory, macroinvertebrates were identified to the lowest feasible taxonomic level. Prior to digestion, organisms were rinsed thoroughly with filtered distilled water to remove externally adhered particles. This step was essential to ensure that only ingested microplastics were quantified, reducing overestimation bias (Hurley et al., 2018).

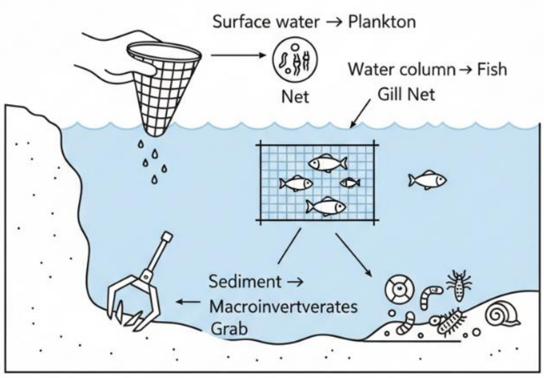

Fig. 3: Sampling strategy for plankton, benthic macroinvertebrates, and fish along the River Sone.

2.2.3 Fish Sampling

Fish specimens were collected using gill nets and cast nets, operated by trained local fishers under ethical guidelines. Only non-threatened and commonly occurring species were selected. Fish were categorized based on trophic guilds (omnivorous, carnivorous, detritivorous) to assess feeding-related differences in microplastic ingestion. Immediately after capture, fish were wrapped in aluminum foil and transported on ice. In the laboratory, total length (L) and body weight (W) were recorded. The gastrointestinal tract (GIT) was dissected using stainless steel instruments. Microplastic ingestion rate (IR) was calculated as:

$$IR = \frac{N_{MP}}{W}$$

where N_{MP} is the number of microplastic particles and W is fish body weight (g), allowing standardized interspecies comparison (Roch et al., 2020).

2.3 Microplastic Extraction and Identification

2.3.1 Chemical Digestion of Organic Matter

Microplastic extraction from biological samples followed standardized digestion protocols. Samples were treated with 30% hydrogen peroxide (H_2O_2) to oxidize organic matter according to the reaction:

$$2H_2O_2 \rightarrow 2H_2O + O_2 \uparrow$$

Digestion was performed at 60°C for 24–48 hours until complete degradation of organic tissues was observed.

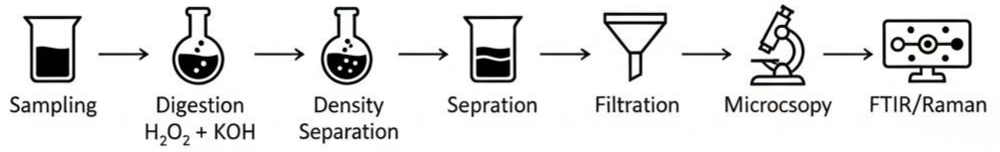

Fig. 4: Analytical workflow for microplastic extraction and polymer identification from freshwater biota.

Hydrogen peroxide was selected due to its efficiency in digesting biological material while preserving polymer integrity (Foekema et al., 2013). Procedural blanks were processed alongside samples to monitor laboratory contamination.

2.3.2 Density Separation

Following digestion, density separation was conducted to isolate microplastics from inorganic residues. Initially, saturated sodium chloride (NaCl) solution (density ≈ 1.20 g cm^{-3}) was used to recover low-density polymers such as polyethylene (PE; density ≈ 0.91–0.96 g cm^{-3}) and polypropylene (PP; density ≈ 0.85–0.92 g cm^{-3}).

Table 2. Physical and chemical properties of polymers relevant to density separation

Polymer	**Chemical Formula**	**Density (g cm^{-3})**	**Separation Medium**
PE	$(C_2H_4)_n$	0.91–0.96	NaCl
PP	$(C_3H_6)_n$	0.85–0.92	NaCl
PET	$(C_{10}H_8O_4)_n$	~1.38	$ZnCl_2$
PVC	$(C_2H_3Cl)_n$	1.3–1.45	$ZnCl_2$

To extract higher-density polymers such as polyethylene terephthalate (PET; density ≈ 1.38 g cm^{-3}) and polyvinyl chloride (PVC; density ≈ 1.30–1.45 g cm^{-3}), zinc chloride ($ZnCl_2$) solution (density ≈ 1.6–1.7 g cm^{-3}) was employed. Separation efficiency (η) was calculated as:

$$\eta = \frac{N_{\text{recovered}}}{N_{\text{total}}} \times 100$$

The supernatant was filtered through glass fiber filters (0.7μ m pore size) and dried in covered glass Petri dishes.

2.3.3 Microscopic and Spectroscopic Identification

Suspected microplastic particles isolated from the samples were initially examined using a stereo microscope at magnifications ranging from 40× to 80× to enable detailed morphological assessment. Particles were systematically categorized according to their shape (fibers, fragments, films, and pellets), size range (from micrometers to millimeters), and visible color. Visual identification was conducted following well-established criteria to minimize misclassification, including the presence of uniform thickness, smooth or angular edges inconsistent with natural materials, the absence of cellular or organic structures, and resistance to deformation or breakage under gentle mechanical pressure. These criteria helped distinguish synthetic particles from natural debris such as plant fibers, shells, or mineral fragments.

To ensure analytical accuracy, the polymeric nature of visually identified particles was subsequently confirmed using Fourier Transform Infrared (FTIR) spectroscopy and Raman spectroscopy. The obtained spectra were carefully compared with standardized reference libraries to identify characteristic absorption bands specific to common polymers, such as polyethylene (PE) exhibiting C–H stretching around ~2915 cm^{-1}, polypropylene (PP) showing CH_3 bending near ~1455 cm^{-1}, and polystyrene (PS) characterized by aromatic C=C stretching at approximately ~1600 cm^{-1}. Only those particles that were conclusively verified through spectroscopic analysis were included in the final microplastic counts, thereby ensuring robustness, reproducibility, and alignment with internationally accepted protocols for microplastic identification (Löder & Gerdts, 2015).

2.4 Quality Assurance and Contamination Control

Strict contamination prevention measures were adopted. Cotton laboratory coats, nitrile gloves, and glass equipment were used throughout. Airborne blanks and reagent blanks were included, and background contamination (B) was corrected using:

$$N_{\text{corrected}} = N_{\text{observed}} - B$$

ensuring data reliability (Masura et al., 2015).

Table 3. Mathematical indices applied for quantifying microplastic ingestion and transfer

Parameter	Equation	Description
MP concentration	$C = N/V$	MPs per m^3
Ingestion rate	$IR = N/W$	MPs per g body weight
Separation efficiency	$\eta = (N_r/N_t) \times 100$	Recovery %
Trophic transfer	$TTF = MP_c/MP_p$	Transfer intensity

Microplastic abundance was expressed as items per individual (fish and macroinvertebrates) and items per cubic meter (plankton). Bioaccumulation across trophic levels was assessed using a trophic transfer factor (TTF):

$$TTF = \frac{MP_{\text{consumer}}}{MP_{\text{prey}}}$$

Statistical analyses were conducted with significance set at $p < 0.05$.

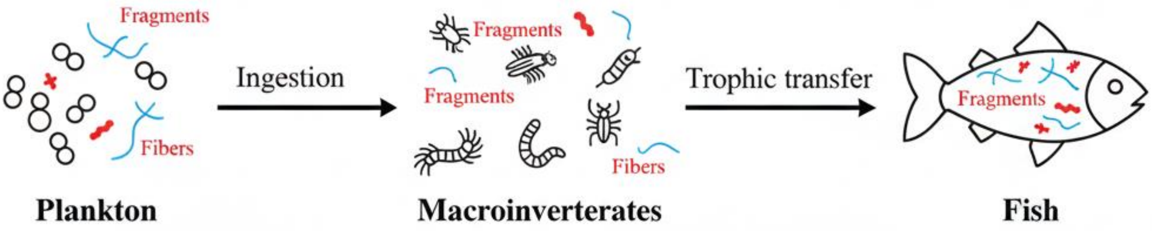

Fig. 5. Conceptual framework illustrating ingestion and trophic transfer of microplastics across freshwater food web of the River Sone.

3. Results

3.1 Occurrence and Abundance of Microplastics Across Trophic Levels

Microplastics were detected in all examined biological compartments of the River Sone ecosystem, including plankton, benthic macroinvertebrates, and fish species, confirming their widespread distribution across freshwater trophic levels. The overall abundance of microplastics increased progressively from lower to higher trophic organisms, indicating effective trophic transfer within the riverine food web. Plankton samples exhibited the lowest microplastic abundance, with concentrations ranging from 2.1–6.4 particles m^{-3} of filtered water. Both phytoplankton and zooplankton fractions contained microplastics, though ingestion was more prominent in zooplankton taxa such as Daphnia spp. and copepods. The relatively lower abundance at this trophic level suggests passive ingestion from the water column rather than bioaccumulation.

In contrast, benthic macroinvertebrates showed significantly higher microplastic loads, ranging between 3.8–12.6 particles $individual^{-1}$, depending on taxa and sampling location. Deposit feeders and detritivores such as chironomid larvae and oligochaetes contained higher microplastic concentrations than predatory macroinvertebrates, highlighting the role of sediments as a major microplastic sink.

Table 4. Microplastic abundance across trophic levels in the River Sone ecosystem

Trophic Level	Sampling Site	Mean Abundance	Unit	Range	Standard Deviation
Plankton	Dehri-on-Sone	3.4	particles m^{-3}	1.8–5.6	±1.1
Plankton	Koilwar	5.9	particles m^{-3}	3.2–7.8	±1.4
Macroinvertebrates	Dehri-on-Sone	5.7	particles $individual^{-1}$	3–9	±1.9
Macroinvertebrates	Koilwar	10.8	particles $individual^{-1}$	7–15	±2.6
Fish	Dehri-on-Sone	11.6	particles $individual^{-1}$	6–18	±3.8
Fish	Koilwar	24.3	particles $individual^{-1}$	15–36	±6.2

Fish species exhibited the highest abundance and diversity of microplastics, with concentrations ranging from 7.2–28.9 particles $individual^{-1}$. Omnivorous and benthivorous fish showed greater microplastic loads than pelagic planktivorous species, supporting the hypothesis that dietary habits strongly influence ingestion rates. These findings collectively indicate a trophic amplification trend from plankton to fish. Similar trophic-level gradients in freshwater ecosystem supporting the robustness of the observed pattern.

3.2 Spatial Distribution of Microplastics

Spatial analysis revealed distinct differences between the midstream site (Dehri-on-Sone) and the downstream site (Koilwar). Downstream samples consistently showed higher microplastic abundance across all trophic levels. Fish collected from Koilwar exhibited up to 35–40% higher microplastic loads compared to those from Dehri-on-Sone. This spatial variation corresponds to increased anthropogenic activities near Koilwar, including urban runoff, fishing activities, domestic wastewater discharge, and agricultural inputs. Macroinvertebrates inhabiting fine sediment zones downstream also showed elevated microplastic accumulation, reinforcing the role of sediment dynamics in microplastic retention.

These results align with earlier studies highlighting rivers as microplastic accumulation corridors, particularly in downstream and urban-influenced stretches (Eerkes-Medrano et al., 2019).

3.3 Morphological Characteristics of Microplastics

Across all biological samples, fibers were the dominant microplastic type, followed by fragments and films. In plankton samples, fibers accounted for approximately 62–70% of total microplastics, while fragments constituted 20–25%, and films were comparatively rare. Benthic macroinvertebrates exhibited a higher proportion of fragments (35–42%), likely due to prolonged exposure to degraded plastic debris in sediments. Fish gut samples showed the greatest morphological diversity, including fibers, fragments, films, and occasional microbeads. Fibers remained dominant (55–60%), particularly in species feeding near the sediment–water interface.

Table 5. Morphological types and size distribution of ingested microplastics

Trophic Level	**Fibers (%)**	**Fragments (%)**	**Films (%)**	**Microbeads (%)**	**Dominant Size Class**
Plankton	68.4	22.7	6.3	2.6	50–300 µm
Macroinvertebrates	54.1	38.6	5.9	1.4	100–1000 µm
Fish	57.8	29.4	9.1	3.7	200 µm–4.5 mm

The dominance of fibrous microplastics is consistent with the prevalence of synthetic textiles, fishing nets, and ropes in freshwater environments and has been widely reported in riverine studies (Li et al., 2021).

3.4 Size Distribution of Ingested Microplastics

Microplastic size analysis revealed that particles <1 mm were most frequently ingested across all trophic levels. Plankton primarily contained microplastics in the 50–300 µm range, which is consistent with the particle sizes available in the water column and within the ingestion limits of zooplankton. Macroinvertebrates showed a broader size distribution, with particles ranging from 100 µm to 2 mm, reflecting sediment exposure and selective feeding behavior. Fish exhibited the widest size range of ingested microplastics (200 µm–4.5 mm), including elongated fibers exceeding 3 mm in length.

Smaller particles are more bioavailable and may translocate across gut epithelium, increasing toxicological risks (Wright & Kelly, 2017). The predominance of smaller particles also suggests continuous fragmentation of plastic debris within the river system.

3.5 Polymer Composition of Microplastics

FTIR and Raman spectroscopic analyses identified polyethylene (PE), polypropylene (PP), and polystyrene (PS) as the dominant polymer types across all trophic levels. Polyethylene accounted for approximately 38–45% of total identified polymers, followed by polypropylene (28–32%) and polystyrene (12–18%).

Table 6. Polymer composition of microplastics identified by FTIR and Raman spectroscopy

Polymer Type	**Plankton (%)**	**Macroinvertebrates (%)**	**Fish (%)**	**Probable Source**
Polyethylene (PE)	42.6	39.1	44.3	Packaging films, carry bags
Polypropylene (PP)	31.2	29.4	27.8	Food containers, ropes
Polystyrene (PS)	14.8	16.6	15.2	Disposable items
Polyethylene terephthalate (PET)	7.1	9.3	8.9	Bottles, textiles
Polyvinyl chloride (PVC)	4.3	5.6	3.8	Pipes, industrial waste

Minor polymer types included PET and PVC, detected mainly in fish and macroinvertebrate samples. The dominance of low-density polymers such as PE and PP reflects their widespread use in packaging materials and their high buoyancy, facilitating dispersion within river systems. These polymer profiles closely match findings from other Indian and global freshwater studies (Mani et al., 2015; Kumar et al., 2022), indicating common sources and transport mechanisms.

3.6 Seasonal Variation in Microplastic Ingestion

Seasonal analysis demonstrated significant variation in microplastic ingestion across sampling periods. Pre-monsoon and post-monsoon seasons showed higher microplastic abundance in all organisms compared to the monsoon season.

During the monsoon, dilution effects due to increased water discharge likely reduced microplastic concentrations in the water column. Post-monsoon samples showed elevated levels, suggesting resuspension of settled microplastics and enhanced feeding activity following monsoon disturbances.

Table 7. Seasonal variation in microplastic ingestion across trophic levels

Season	Plankton (particles m^{-3})	Macroinvertebrates (particles individual^{-1})	Fish (particles individual^{-1})	Statistical Significance (ANOVA)
Pre-monsoon	5.8 ± 1.3	10.9 ± 2.4	26.1 ± 5.9	$p < 0.01$
Monsoon	2.6 ± 0.9	5.1 ± 1.7	12.3 ± 3.4	$p < 0.05$
Post-monsoon	4.9 ± 1.1	9.4 ± 2.1	22.7 ± 4.8	$p < 0.01$

Fish collected during the pre-monsoon period exhibited the highest microplastic loads, potentially due to reduced water flow, higher residence time of pollutants, and increased feeding intensity.

3.7 Evidence of Trophic Transfer and Bioaccumulation

The progressive increase in microplastic abundance from plankton to macroinvertebrates and fish provides strong evidence of trophic transfer within the River Sone food web. While plankton serve as the primary entry point, macroinvertebrates act as intermediate reservoirs, facilitating transfer to higher trophic levels. Fish gut analyses revealed intact fibers and fragments, indicating limited egestion and potential for prolonged gut retention. Although this study focused on gut contents, the observed accumulation patterns suggest the possibility of pseudo-bioaccumulation, where continuous ingestion exceeds egestion rates.

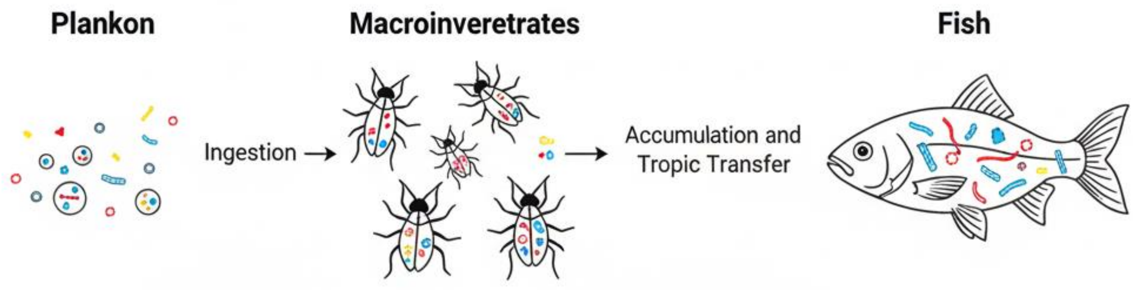

Fig. 6: Schematic Representation of Microplastic trophic transfer

Such trophic transfer pathways have been demonstrated experimentally and observationally in freshwater and marine systems (Setälä et al., 2014; Nelms et al., 2018), raising concerns regarding ecological and human health implications.

4. Discussion

The present study provides comprehensive evidence of microplastic contamination across multiple trophic levels in the River Sone ecosystem, highlighting the pervasive nature of plastic pollution in freshwater environments of developing regions. The detection of microplastics in plankton confirms their early entry point into the aquatic food web, consistent with the role of plankton as primary consumers and key vectors for contaminant transfer. As plankton indiscriminately filter particles within specific size ranges, microplastics present in the water column are readily ingested, facilitating their movement into higher trophic levels. The observed increase in microplastic abundance from plankton to benthic macroinvertebrates and fish strongly suggests trophic transfer through both predation and detrital feeding pathways. Macroinvertebrates, particularly deposit feeders, exhibited elevated microplastic loads, reflecting prolonged exposure to contaminated sediments. Sediments are known to act as sinks for microplastics, especially for denser or biofouled particles, thereby increasing ingestion risk for benthic organisms. This intermediate trophic level thus functions as a critical reservoir, enabling the onward transfer of microplastics to fish.

Fish demonstrated the highest abundance and diversity of ingested microplastics, which can be attributed to their longer lifespan, broader dietary spectrum, and cumulative exposure. Benthivorous and omnivorous fish showed particularly high microplastic loads, supporting the hypothesis that dietary habits significantly influence ingestion rates. Similar trophic amplification patterns have been reported in freshwater and marine ecosystems, where repeated ingestion through contaminated prey results in pseudo-bioaccumulation of microplastics in higher trophic organisms. The dominance of fibrous microplastics across all trophic levels is a notable finding and aligns with global observations in freshwater systems. Synthetic fibers primarily originate from domestic wastewater, laundering of synthetic textiles, fishing gear degradation, and rope materials. Given the limited wastewater treatment infrastructure in many parts of Bihar, untreated or partially treated effluents likely serve as a major source of fibrous microplastics entering the River Sone. Fibers are particularly concerning due to their elongated shape, which can enhance gut retention time and increase the potential for physical abrasion and blockage.

Polymer analysis revealed the predominance of polyethylene, polypropylene, and polystyrene, reflecting widespread use of single-use plastics and packaging materials. These low-density polymers are easily transported within river systems and can remain bioavailable for extended periods. Their presence across all trophic levels indicates persistent environmental exposure and highlights rivers as efficient conduits for plastic transport and ecological interaction. Comparable polymer profiles have been reported in other Indian rivers, including the Ganga and Yamuna, suggesting common sources and pathways of plastic pollution. Seasonal variation in microplastic ingestion observed in this study underscores the influence of hydrological conditions on microplastic distribution and bioavailability. Lower microplastic abundance during the monsoon season is likely due to dilution effects and increased water discharge, which reduce particle residence time in the water column. In contrast, higher ingestion during pre-monsoon and post-monsoon periods may result from reduced flow velocity, enhanced sediment resuspension, and increased feeding activity. Post-monsoon resuspension of settled microplastics has been identified as a key mechanism driving seasonal peaks in contamination.

Although this study focused on gut content analysis, continuous ingestion of microplastics raises concerns regarding sub-lethal physiological stress in aquatic organisms. Laboratory and field studies have demonstrated that microplastics can induce oxidative stress, inflammation, altered energy metabolism, and impaired growth and reproduction in freshwater biota. The microplastics can act as vectors for toxic additives, heavy metals, and pathogenic microorganisms, further amplifying their ecological impact. From an ecological perspective, the trophic transfer of microplastics may alter feeding efficiency, energy flow, and predator–prey interactions, potentially leading to long-term ecosystem-level effects. In river systems such as the Sone, which support local fisheries and human consumption, the accumulation of microplastics in fish also raises food safety and public health concerns. While the direct health implications of dietary microplastic exposure in humans remain under investigation, the presence of microplastics in edible fish tissues warrants precautionary monitoring.

The findings of this study highlight the River Sone as a vulnerable freshwater ecosystem subject to significant microplastic stress, driven by anthropogenic activities and hydrological dynamics. The clear evidence of trophic transfer emphasizes the need for integrated management strategies, including improved wastewater treatment, plastic waste reduction, and long-term ecological monitoring. Future research should focus on tissue translocation, toxicological responses, and risk assessment frameworks to better understand the implications of microplastics for freshwater biodiversity and human health.

5. Conclusion

This study presents the first comprehensive evidence of microplastic ingestion and trophic transfer within the freshwater food web of the River Sone, an ecologically and socio-economically important tributary of the Ganga River system. The detection of microplastics in plankton confirms their early entry into the aquatic ecosystem, while the progressively higher abundance observed in benthic macroinvertebrates and fish provides strong empirical support for trophic transfer and accumulation across successive trophic levels. These findings underscore the growing vulnerability of freshwater food webs in densely populated and anthropogenically influenced river basins of India. The predominance of synthetic fibers and low-density polymers such as polyethylene and polypropylene across all biological compartments reflects widespread plastic usage and inadequate waste and wastewater management practices in the region. Seasonal variability in microplastic ingestion further highlights the role of hydrological processes, particularly river discharge, sediment resuspension, and pollutant residence time, in governing microplastic bioavailability. Together, these results indicate that microplastic contamination in riverine ecosystems is not static but dynamically influenced by both natural and human-induced factors. The focused on gut content analysis, the continuous ingestion of microplastics by aquatic organisms raises important concerns regarding sub-lethal physiological stress, ecological imbalance, and long-term biodiversity impacts. The accumulation of microplastics in fish, many of which are consumed by local communities, also signals potential implications for food safety and human health, warranting precautionary attention. Rivers such as the Sone thus function not only as transport pathways for plastic debris but also as active ecological systems where plastics interact with biota and food webs.

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