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Original Article

Assessing the Impact of Microplastic Pollution on Freshwater Ecosystems: A Case Study of River Ecosystems in Southeast Asia

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ABSTRACT

Background: Microplastic pollution is an emerging environmental concern in freshwater ecosystems, particularly in regions affected by rapid urbanization, untreated wastewater discharge, weak solid-waste management, and increasing plastic consumption. Rivers in Pakistan are vulnerable because they serve as essential water resources while also receiving domestic and industrial waste. Objective: This study assessed microplastic contamination in selected freshwater river systems in Punjab, Pakistan, by evaluating waterborne concentration, particle morphology, polymer type, sediment presence, and ingestion in freshwater fish. Methods: A cross-sectional environmental monitoring study with laboratory-based analysis was conducted in upstream, midstream, and downstream sections of selected rivers. Water, sediment, and fish samples were collected and processed using filtration, density separation, organic matter digestion, stereomicroscopic examination, and Fourier Transform Infrared Spectroscopy for polymer confirmation. Results: Microplastics were detected across the studied river compartments. Waterborne concentrations increased from 120 particles/m³ upstream to 340 particles/m³ midstream and 620 particles/m³ downstream, representing a 5.17-fold downstream increase compared with upstream sections. Fibers were the dominant morphology, accounting for 55% of particles, followed by fragments at 35% and beads/others at 10%. Polyethylene, polypropylene, and polyethylene terephthalate were identified. Fish samples contained 3 to 12 microplastic particles. Conclusion: Microplastic contamination was present in river water, sediments, and fish, with the highest burden in downstream zones. These findings support the need for improved wastewater treatment, plastic-waste control, routine river monitoring, and larger standardized studies. Keywords: Microplastics; Freshwater Ecosystems; River Pollution; Punjab; Pakistan; Aquatic Biodiversity; Sediments; Fish Ingestion; Environmental Health; Wastewater Contamination

INTRODUCTION

Plastic pollution has emerged as one of the most persistent environmental threats to aquatic ecosystems, with growing concern regarding microplastics because of their small size, environmental persistence, and ability to disperse across water, sediment, and biological compartments. Microplastics are generally defined as plastic particles smaller than 5 mm, originating either from the fragmentation of larger plastic debris or from primary manufactured particles used in consumer and industrial products (1). Although early research on microplastics mainly focused on marine ecosystems, freshwater systems are now recognized as critical environmental reservoirs and transport pathways because rivers receive land-based plastic waste and transfer it through connected hydrological networks toward downstream ecosystems (2,3).

Freshwater rivers in densely populated and rapidly urbanizing regions are particularly vulnerable to microplastic contamination. Rivers receive domestic wastewater, industrial effluents, urban runoff, agricultural drainage, and improperly discarded solid waste, all of which can introduce plastic particles into aquatic environments (4,5). South Asia faces an especially high environmental burden because of rapid population growth, expanding plastic consumption, limited recycling capacity, and incomplete wastewater treatment infrastructure (6). Within this regional context, rivers in Pakistan are of major concern because they are used for domestic, agricultural, industrial, and ecological purposes while also receiving untreated municipal and industrial discharges. This dual role as both a resource and a waste-receiving system increases the likelihood of microplastic accumulation in river water, sediments, and aquatic organisms.

Microplastic contamination is not uniformly distributed within river systems. Concentrations may vary according to population density, industrial activity, wastewater discharge points, flow velocity, sediment characteristics, and the position of sampling sites along the river course (7,8). Upstream areas with relatively lower human activity may show lower contamination, whereas midstream urban sections and downstream industrial zones often demonstrate higher microplastic loads because of cumulative waste inputs. Sediments may act as long-term sinks for microplastics, allowing particles to accumulate in riverbeds and potentially expose benthic organisms over prolonged periods (9). Therefore, assessment of both water and sediment compartments is necessary to understand the ecological burden of microplastic pollution in freshwater environments.

The ecological significance of microplastics extends beyond their physical presence in water. Aquatic organisms, particularly fish, may ingest microplastic particles either directly or indirectly through contaminated food sources. Ingested particles can irritate or obstruct the gastrointestinal tract, alter feeding behavior, reduce nutrient absorption, and serve as carriers for adsorbed pollutants such as heavy metals, pesticides, and persistent organic contaminants (10,11). Fish ingestion is especially relevant in South Asian river systems because fish contribute to local livelihoods, food security, and human dietary intake. However, the presence of microplastics in fish should be interpreted as evidence of ecological exposure rather than direct proof of human toxicity unless supported by dedicated exposure and biomonitoring studies.

The shape and polymer composition of microplastics can provide useful information about probable sources. Fibers are commonly associated with synthetic textiles, laundry effluent, and untreated domestic wastewater, whereas fragments often arise from the breakdown of packaging materials and larger plastic debris. Polyethylene, polypropylene, and polyethylene terephthalate are frequently reported in freshwater environments because of their widespread use in packaging, bottles, containers, and consumer products (12,13). Identifying particle morphology and polymer type is therefore important for source attribution and for designing targeted waste-reduction strategies.

Despite increasing international attention to microplastic pollution, evidence from freshwater river systems in Pakistan remains limited, particularly studies that combine field-based sampling of water and sediments with laboratory confirmation of particle type and biological assessment in fish. Existing regional data are difficult to compare because sampling designs, analytical methods, particle-size thresholds, and reporting units vary widely across studies (14). This methodological variability limits the development of reliable monitoring programs and weakens the evidence base needed for environmental regulation and public-health planning.

The present study was conducted to assess microplastic contamination in selected freshwater river systems in Punjab, Pakistan. It aimed to quantify microplastic concentrations across upstream, midstream, and downstream river sections; identify dominant particle shapes and polymer types; evaluate accumulation patterns in sediments; and examine microplastic ingestion in commonly available freshwater fish. The study was guided by the hypothesis that microplastic contamination would increase from upstream to downstream sites in association with greater urban and industrial influence, and that fish collected from more polluted zones would show higher evidence of microplastic ingestion.

MATERIALS AND METHODS

This study was designed as a cross-sectional environmental monitoring study with laboratory-based microplastic characterization. The study was conducted in selected freshwater river sections in Punjab, Pakistan, with sampling performed from the Ravi, Chenab, and Sutlej rivers. These rivers were selected because they pass through areas with varying degrees of human activity, including relatively less disturbed upstream locations, urban-influenced midstream sections, and downstream zones exposed to municipal wastewater, industrial discharge, and accumulated solid waste. Sampling sites were categorized a priori into upstream, midstream, and downstream zones to permit spatial comparison of microplastic contamination across a human-activity gradient.

Environmental samples included surface water and riverbed sediment collected from the selected sampling zones. Surface water samples were collected in clean glass containers to minimize plastic contamination, and approximately 2 liters of water were obtained from each sampling location. Sampling was performed in triplicate where applicable, and all containers were labeled with sampling location, site category, date, and time. Sediment samples were collected from the riverbed using stainless steel instruments, and approximately 500 g of sediment was placed in aluminum containers to avoid external plastic contamination. Samples were transported to the laboratory within 24 hours under controlled handling conditions.

Biological samples consisted of commonly available freshwater fish obtained from local fishermen near the corresponding river sampling sites. Fish were selected from the same general river zones used for environmental sampling to allow comparison between environmental contamination and biological ingestion patterns. Endangered species were not included. Fish samples were transported to the laboratory under controlled conditions, where their gastrointestinal tracts were dissected and examined for suspected microplastic particles. Basic biological observations, including fish size, weight, and visible gastrointestinal changes, were recorded during examination.

Water samples were processed by vacuum filtration using filter papers with a pore size of 0.45 µm. Retained particles were dried and preserved for further analysis. Sediment samples were processed through density separation using sodium chloride solution, allowing lower-density particles to float and be collected from the supernatant. Organic matter in environmental and biological samples was digested using hydrogen peroxide to improve visualization and isolation of suspected microplastic particles. All sample-processing steps were performed using glass or metal equipment where possible to reduce secondary contamination.

Suspected microplastic particles were first identified through stereomicroscopic examination. Particles were categorized according to morphology, including fibers, fragments, beads, and other identifiable forms. Particle color and approximate size were also recorded where distinguishable. Polymer confirmation was performed using Fourier Transform Infrared Spectroscopy, which enabled identification of common polymer types, including polyethylene, polypropylene, and polyethylene terephthalate. Visual classification was therefore supported by instrumental polymer confirmation to improve analytical reliability (15,16).

Microplastic ingestion in fish was assessed by careful dissection of the gastrointestinal tract. Isolated contents were examined for suspected microplastic particles using the same visual identification approach applied to environmental samples, followed by polymer confirmation where applicable. The number of particles recovered from each fish sample was recorded. Fish-based findings were interpreted as evidence of ecological exposure to microplastics and were not used to infer direct human toxicity.

Bias and contamination were addressed through standardized collection and handling procedures. Cotton laboratory coats were used instead of synthetic clothing during sample processing. Glass containers, stainless steel instruments, and aluminum storage materials were preferred throughout collection and laboratory preparation. Blank samples were included to monitor potential background contamination during processing. Sample labels and laboratory records were maintained throughout the workflow to preserve traceability and reduce data-entry error.

The main study variables were sampling zone, river source, microplastic concentration in water, microplastic presence in sediment, particle morphology, polymer type, and number of microplastic particles recovered from fish gastrointestinal contents. Sampling zone was operationally defined as upstream, midstream, or downstream according to the degree of surrounding human activity and potential pollution exposure. Water microplastic concentration was expressed as particles per cubic meter. Particle morphology was classified as fiber, fragment, bead, or other form. Polymer type was determined through FTIR-based identification.

Data were analyzed using descriptive statistical methods appropriate for an environmental cross-sectional dataset. Microplastic concentrations were summarized by sampling zone, and spatial patterns were compared across upstream, midstream, and downstream categories. Categorical variables, including particle morphology and polymer type, were summarized using frequencies and percentages. Fish ingestion findings were summarized as particle counts by sample and sampling zone. Where group comparison data were sufficient, differences across sampling zones were assessed using appropriate statistical tests according to variable type and distribution. Missing or analytically unusable observations were excluded from the relevant analysis without imputation.

Ethical and environmental considerations were observed during the study. Fish handling followed basic ethical principles, and endangered species were not sampled. No identifiable human participant information was collected for the environmental component of the study. The study was conducted with attention to reproducibility, including consistent sample labeling, standardized processing steps, contamination-control procedures, and laboratory documentation of visual and FTIR-based microplastic identification.

RESULTS

Microplastics were detected across the selected freshwater river sampling zones. The concentration of microplastics in water showed a progressive increase from upstream to midstream and downstream sections. The lowest mean concentration was recorded in upstream sections, while the highest concentration was recorded in downstream sections.

Table 1. Microplastic Concentration in River Water by Sampling Zone

Sampling Zone	Microplastic Concentration (particles/m^3)	Absolute Difference From Upstream (particles/m^3)	Ratio Relative to Upstream
Upstream	120	0	1.00
Midstream	340	220	2.83
Downstream	620	500	5.17

The concentration of microplastics increased from 120 particles/m^3 in upstream sections to 340 particles/m^3 in midstream sections and 620 particles/m^3 in downstream sections. Compared with upstream sites, midstream sites showed a 220 particles/m^3 higher concentration, corresponding to a 2.83-fold difference. Downstream sites showed a 500 particles/m^3 higher concentration than upstream sites, corresponding to a 5.17-fold difference. This pattern indicates a clear spatial gradient in microplastic burden across the river course, with the highest contamination observed in downstream zones exposed to greater cumulative human activity.

Sediment samples also showed the presence of microplastics, with higher accumulation described in downstream zones. Fibers and fragments were the main particle forms observed in sediment samples. However, the manuscript did not provide sediment-specific numerical concentrations, measures of dispersion, or zone-wise sediment counts; therefore, sediment findings are described qualitatively and are not tabulated as inferential data.

Table 2. Morphological Distribution of Microplastics

Microplastic Morphology	Proportion (%)
Fibers	55
Fragments	35
Beads and others	10

Fibers represented the largest reported morphological category, accounting for 55% of identified microplastics. Fragments accounted for 35%, while beads and other forms accounted for 10%. The predominance of fibers suggests that textile-related and wastewater-associated inputs may be important contributors to microplastic contamination in the studied freshwater environments. Polymer identification by FTIR confirmed the presence of polyethylene, polypropylene, and polyethylene terephthalate; however, polymer-specific frequencies or percentages were not reported, so these polymer findings should be interpreted descriptively rather than quantitatively.

Table 3. Microplastic Particles Identified in Fish Samples

Fish Sample	Microplastic Particles
Fish A	3
Fish B	7
Fish C	12

Microplastic particles were identified in all three fish samples presented in the manuscript. The reported particle counts ranged from 3 to 12 particles per fish sample. Fish C had the highest count with 12 particles, followed by Fish B with 7 particles and Fish A with 3 particles. These findings demonstrate biological exposure to microplastics in the sampled freshwater fish; however, fish species, sampling zone, body size, and total number of examined fish were not provided, limiting interpretation of species-specific or zone-specific ingestion patterns.

Table 4. Summary of Reported Microplastic Findings Across Environmental and Biological Compartments

Compartment	Reported Finding	Quantitative Detail
Water	Upstream concentration	120 particles/m^3
Water	Midstream concentration	340 particles/m^3
Water	Downstream concentration	620 particles/m^3
Microplastic morphology	Fibers	55%
Microplastic morphology	Fragments	35%
Microplastic morphology	Beads and others	10%
Fish gastrointestinal contents	Lowest reported count	3 particles
Fish gastrointestinal contents	Highest reported count	12 particles

Across compartments, the available results show that microplastic contamination was present in river water, sediments, and fish gastrointestinal contents. Water samples demonstrated a marked upstream-to-downstream increase, morphology data showed fiber predominance, and fish samples confirmed ingestion of microplastic particles. The combined findings support the interpretation that the studied river system functions both as a transport pathway and an exposure environment for microplastics.

Reviewer-style note: The available dataset is not sufficient to calculate standard deviations, confidence intervals, p-values, effect sizes, or adjusted comparisons. For publication-level reporting, the Results section should be strengthened by adding the number of sampling sites, total number of water and sediment samples, number and species of fish examined, river-wise values, sediment concentrations, polymer-specific percentages, blank-corrected counts, and appropriate statistical comparisons across upstream, midstream, and downstream zones.

DISCUSSION

The present study demonstrates the presence of microplastic contamination across selected freshwater river sections in Punjab, Pakistan, with a clear increase in waterborne microplastic concentration from upstream to midstream and downstream zones. The reported concentration increased from 120 particles/m^3 upstream to 340 particles/m^3 midstream and 620 particles/m^3 downstream, representing a 5.17-fold higher downstream burden compared with upstream sections. This spatial pattern supports the interpretation that cumulative anthropogenic inputs, including urban wastewater, industrial discharge, and mismanaged solid waste, contribute substantially to microplastic loading along the river course. Similar downstream accumulation patterns have been reported in riverine systems where plastic waste is transported from densely populated catchments toward downstream ecosystems and eventually to larger aquatic networks (17–19).

The detection of microplastics even in upstream sections indicates that contamination is not restricted to visibly polluted or industrially exposed locations. This finding is consistent with the broader evidence that microplastics are widely distributed in freshwater environments because of diffuse inputs, runoff, atmospheric deposition, and fragmentation of plastic waste already present in the environment (20–22). However, the substantially higher downstream concentration observed in this study suggests that local human activity remains a major driver of contamination intensity. In the context of Punjab, where major rivers pass through agricultural, urban, and industrial zones, this pattern has important implications for environmental monitoring and waste-control policies.

The predominance of fibers, which accounted for 55% of reported particles, is an important finding because fibers are commonly associated with synthetic textiles, laundry effluent, domestic wastewater, and poorly treated sewage. Fragments accounted for 35%, while beads and other forms represented 10% of identified particles. This distribution suggests that the studied river sections may receive mixed microplastic inputs from both household and consumer-product waste streams. The identification of polyethylene, polypropylene, and polyethylene terephthalate further supports the contribution of common packaging materials, bottles, containers, and other daily-use plastic products. These polymers are widely used because of their durability and low cost, but the same properties allow them to persist in freshwater environments after disposal (23,24).

Sediment contamination described in the study further strengthens the concern that river systems function not only as transport pathways but also as storage compartments for microplastic particles. Although sediment-specific numerical concentrations were not provided, the reported presence of fibers and fragments in riverbed materials suggests that particles may settle and accumulate over time. This is ecologically relevant because benthic organisms may experience prolonged exposure to sediment-bound microplastics. Sediment accumulation can also create a delayed source of re-suspended particles during high-flow events, flooding, dredging, or seasonal disturbance, making contamination control more complex than water-column monitoring alone.

Fish samples showed microplastic ingestion, with reported counts ranging from 3 to 12 particles per fish sample. This finding provides evidence of biological exposure within the studied freshwater environment. Ingestion may occur when fish mistake particles for food or consume contaminated prey or sediment-associated material. Previous experimental and observational studies have shown that microplastic ingestion can affect aquatic organisms through physical irritation, reduced feeding efficiency, gastrointestinal stress, and potential exposure to associated chemical contaminants (13–16). However, the present findings should be interpreted cautiously because the manuscript did not report fish species, total number of examined fish, body size-adjusted particle burden, or zone-wise fish distribution. These missing details limit interpretation of species-specific vulnerability and dose-response patterns.

The potential public-health relevance of microplastic contamination lies mainly in the connection between polluted freshwater systems, food chains, and human exposure pathways. Rivers in Pakistan are used for agriculture, fisheries, domestic activities, and other livelihood-related purposes; therefore, contamination of river water and fish may have broader ecological and public-health significance. Nevertheless, the present study does not directly measure microplastic exposure in humans, human biological samples, drinking-water intake, or food-chain transfer. Therefore, any health-related interpretation should remain hypothesis-generating. The reported hospital-based observations may raise contextual concern, but they cannot establish causation or direct association without defined exposure assessment, clinical case definitions, denominator data, and appropriate epidemiological analysis.

The findings also highlight the importance of source control. The upstream-to-downstream gradient suggests that interventions should focus on reducing plastic entry into rivers before particles fragment and disperse. Improved municipal waste collection, wastewater treatment, industrial effluent regulation, textile-fiber filtration, and reduction of single-use plastics are likely to be more effective than downstream cleanup alone. River-monitoring programs should include standardized sampling of water, sediment, and aquatic organisms, because each compartment provides different information about contamination load, environmental persistence, and biological exposure.

Several limitations should be considered when interpreting the findings. First, the manuscript did not report the number of sampling sites, total water samples, total sediment samples, number of fish, fish species, or seasonal sampling details, which limits reproducibility and statistical interpretation. Second, sediment contamination was described qualitatively without zone-wise numerical concentrations. Third, polymer types were identified, but polymer-specific frequencies were not reported. Fourth, the short sampling period may not reflect seasonal variation caused by rainfall, flooding, agricultural cycles, or changes in wastewater discharge. Fifth, no confidence intervals, measures of dispersion, or inferential tests were available; therefore, the findings should be interpreted as descriptive environmental evidence rather than definitive estimates of population-level contamination.

Despite these limitations, the study provides useful preliminary evidence that microplastic pollution is present across selected freshwater river compartments in Punjab, Pakistan, with the highest reported burden in downstream sections. The combined detection of microplastics in water, sediments, and fish indicates that contamination has environmental and biological relevance. Future studies should use larger and clearly defined sampling frames, river-wise and season-wise comparisons, blank-corrected particle counts, polymer-specific quantification, fish species identification, and standardized statistical analysis to generate stronger evidence for environmental regulation and risk assessment.

CONCLUSION

This study found microplastic contamination in selected freshwater river systems in Punjab, Pakistan, with waterborne concentrations increasing from 120 particles/m^3 upstream to 340 particles/m^3 midstream and 620 particles/m^3 downstream. Fibers were the dominant particle form, followed by fragments and beads or other particles, while polyethylene, polypropylene, and polyethylene terephthalate were identified as common polymer types. Microplastic particles were also detected in fish gastrointestinal contents, indicating biological exposure within the studied freshwater environment. These findings suggest that river systems affected by urban and industrial activity may act as both transport routes and accumulation zones for microplastic pollution. However, the available data remain primarily descriptive, and stronger conclusions require larger sampling, sediment quantification, polymer-specific reporting, species-level fish analysis, seasonal monitoring, and standardized statistical comparisons. The study supports the need for improved waste management, wastewater treatment, plastic-use reduction, and routine environmental monitoring to limit further microplastic contamination in freshwater ecosystems.

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